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SOME EARTHWORMS FROM THE SOUTH SEA ISLANDS¹⁾

By

SHINJIRÔ KOBAYASHI

Keijô Second Higher Common School

(With 1 text-figure)

(Received October 18, 1939)

From the South Sea Islands under the Japanese mandate only four species of earthworms have hitherto been reported. They are *Pheretima recta* and *Ph. taitensis* from the Jaluit and Marshall Islands^(1, 11 & 12), and *Ph. carolinensis* and *Drawida barwelli* from the Carolines^(2 & 3).

But Professor TEISÔ ESAKI, of the Kyûshû Imperial University, has recently, in the Mariana and Caroline Islands, collected several other specimens of earthworms, and by his courtesy I have had the opportunity of examining them. They are referable to two species of *Dichogaster bolau*i (MICHAELSEN) and *Pheretima* sp. (juvenile). *D. bolau*i is a species highly variable and is widely distributed in the warmer regions of the globe.

The present report forms the first records of this species from Micronesia.

I wish to express my hearty thanks to Prof. T. ESAKI for the great kindness he has shown me in supplying the material. My most sincere thanks must be extended to Prof. SANJI HÔZAWA of the Tôhoku Imperial University for the kind help he has given me in my study of the specimens.

Family Megascolecidae

Subfam. Octochaetinae

Genus *Dichogaster* BEDDARD

*Dichogaster bolau*i (MICHAELSEN) 1891

1900 *D. bolau*i, MICHAELSEN, Tierreich, Oligochaeta, p. 340 (see this paper - for complete synonymy and bibliography up to 1900). 1901 *D. bolau*i, MICHAELSEN, Bull. Acad. Imp. Sci. St.-Petersburg, XII, 2, p. 205; 1903 MICHAELSEN, Sb. Böhm. Ges. Prag, XL, p. 16; 1910 MICHAELSEN, Abh. Ver. Hamburg, XIX, p. 98; 1913 MICHAELSEN, Nova Caledonia, Zool., I, 5, p. 273; 1913 MICHAELSEN, Mem. Soc. neu. Sci. nat., V, p. 214;

¹⁾ Results of Professor T. ESAKI's Micronesia Expeditions, 1936-1938, No. 30.

1916 MICHAELSEN, Kungl. Sv. Vetenskapsakad. Handl., LII, 13, p. 37; 1916 *D. bolauai palmicola*, STEPHENSON, Rec. Ind. Mus., XII, p. 348; 1917 *D. bolauai*, STEPHENSON, *ibid.*, XIII, p. 413; 1920 STEPHENSON, Mem. Ind. Mus., VII, p. 257; 1922 MICHAELSEN, Capita Zool., I, 3, p. 18; 1923 STEPHENSON, Fauna British India, Oligochaeta, pp. 472-473; 1924 STEPHENSON, Rec. Ind. Mus., XXVI, pp. 132-133; 1925 STEPHENSON, *ibid.*, XXVII, p. 73; 1926 STEPHENSON, *ibid.*, XXVIII, p. 266; 1928 MICHAELSEN, Ark. Zool., 20 A, 3, p. 9; 1931 STEPHENSON, P. Z. S. London, p. 65; 1931 STEPHENSON, Rec. Ind. Mus., XXXIII, pp. 195-197; 1932 PICKFORD, Disc. Rep., IV, pp. 286-287, fig. 2 j-1; 1935 ČERNOSVITOV, Capita Zool., VI, 1, p. 12; 1938 PICKFORD, Carnegie Inst. Washington Pub., 491, pp. 98-99.

External characteristics: Body length is 17 mm in one complete semi-mature specimen and is 14 mm in one specimen mature but with regenerated tail. Greatest diameter in clitellar region $1\frac{1}{3}$ - $1\frac{1}{2}$ mm. Number of segments 94. Colour uniformly buff, unpigmented, but clitellum brownish.

Prostomium proepilobous. Segment I is not distinctly demarcated from II. First dorsal pore in 5/6, distinct and functional; an indistinct and non-functional pore was found in 4/5 in two specimens. No dorsal pores were found on clitellum.

Setae closely paired; setal distance *aa* subequal to, or a little greater than, *bc*; *ab* subequal to *cd*; *dd* subequal to $\frac{2}{3}$ of the circumference being measured in one segment immediately posterior to the clitellum.

Clitellum in XIII-XX, well defined, saddle-shaped, but its ventral part also slightly thickened comparing it with the general surface of the body. Glandularity extends to *a*-line, but in the lateral part it is slightly less developed than the dorsal, the intersegmental furrows being still present there.

Prostatic pores, two pairs in XVII and XIX, on *ab*-line. Seminal groove almost straight; it is slightly concave lateralwards in one semi-mature specimen. Margin of groove is slightly thickened and weakly whitened. Male pores were not identified.

Female pore single, midventrally on XIV, at the centre of an epidermal elevation which is indistinctly demarcated, glandulated and light-coloured.

Spermathecal pores were not visible. But, their position was indirectly recognizable from slight depressions with glandular colouration in margin. Two pores in 7/8 and 8/9, on *ab*-line.

Internal anatomy: Owing to the small size of body it is very difficult to determine the disposition of the septa. But, we are able to interpret it indirectly judging from the situation of spermathecae. None of these septa are especially thickened.

Two gizzards in VII and VIII, rather distinctly constricted. Crop moderately dilated and weakly muscled.

Three pairs of calciferous glands are found of oval shape in XV-XVII. Intestine begins in XVIII. Last pair of hearts in XII. Seminal vesicles rudimentary, in XI (and XII?); in XII vesicle-like fragments only were found.

Spermathecae very small. Ampulla ovoidal or pear-shaped. Duct thick, longer than ampulla, distinctly marked off from the latter. Diverticulum club-shaped, arising from about the middle of the duct.

Penial setae are typical, showing the characteristic dimorphism of the species (Fig. 1). Hooked setae, length $\sim 0.32\text{--}0.35$ mm, thickness $3.8\text{--}4.5\ \mu$ distally, $5.6\text{--}6.1\ \mu$ in the middle part, and $7\text{--}7.6\ \mu$ proximally; scalpel-shaped setae, length $0.26\text{--}0.29$ mm, thickness $3.4\text{--}3.9\ \mu$ distally, $4.8\text{--}5.1\ \mu$ in the middle part, $6\text{--}6.5\ \mu$ proximally. Number of teeth found on a hooked seta is 4 in all of 8 cases examined.

Localities and materials: Lelo, Kusaie, Caroline Isls., Dec. 10, 1937, 1 mature and 3 immature specimens; Sonson - Taipingot, Rota, Mariana Isls., Nov. 6, 1937, 2 semi-mature specimens.

Distribution: Africa, Madagascar, India, Burma, California, Mexico, Central and S. America, W. Indies, Germany (Hamburg), New-Caledonia, Loyalty Isls., Mariana Isls., Caroline Isls., Philippine Isls., Borneo, Malay St., (Malay Archipelago - if *D. malayana* is really synonymous with the present species), Hainan (after the completion of this manuscript, I received a paper of 'Oligochaeta from Hainan, Kwangtung '38' from CHEN).

Remarks: *D. bolau* is a highly variable species, many names having been given to it hitherto. It seems to be certain that the present specimens belong to the pigmy typical form judging from the characteristics seen in the small size of the body, in the arrangement of the nephridia, and in the rudimentary seminal vesicles.

One of the Columbian specimens reported by MICHAELSEN⁽⁵⁾ is only $15\text{ mm} \times 1\frac{1}{2}\text{ mm}$. A complete specimen in the present collection is $17\text{ mm} \times 1\frac{1}{3}\text{--}1\frac{1}{2}\text{ mm}$. These two specimens above mentioned are smaller than the

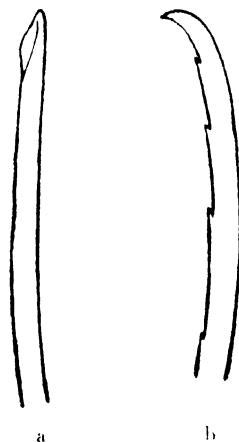


Fig. 1. *Dichogaster bolau*.
a, distal end of a scalpel-shaped seta; b, distal end of a hooked seta. ca. $\times 960$

usual run of the species, 20–40 mm^(1 & 8). According to STEPHENSON⁽⁹⁾ it is 50 mm in the average length of a number of specimens collected from Siju Cave, Assam. (PICKFORD's specimen from Guinea⁽¹⁰⁾ is as large as 69 mm × 2 mm. Possibly as she remarked it may be a variety of the present species.)

Female pore is seated on a glandular epidermal thickening which is not very conspicuous but is easily distinguishable from the neighbouring surface. There was not found any kind of groove, by which the thickening is delimited forming a "papilla"^{1, 9 & 10}. However, that may be owing to the incomplete maturity of the specimen.

There are not any records which have alluded to the presence of the dorsal pore in 4/5. But in the case of the present specimens it is rather indistinct and seems to be not functional.

Penial setae are typical in shape. But, of the two kinds of setae, the hooked one is notably thicker than the scalpel-shaped as in the case of Indian specimens⁽¹⁰⁾. On the other hand these features are reversed in the case of PICKFORD's Yucatan specimens⁽⁷⁾. Number of teeth found on the hooked setae has been generally known to be eight^(1 & 8). But, it was four in all of the cases examined. According to STEPHENSON⁽¹¹⁾, it varies within the range of 4–6 in the cases of Indian specimens. It is four in the Yucatan specimens too.

Clitellum is saddle-shaped, but the ventral part is, however, a little thicker than the general surface of the body. This characteristic as found in this organ has been also recorded by STEPHENSON⁽¹⁰⁾ in the cases of Burmese and Philippine specimens. Mainly from this characteristics, he has discussed the probable synonymy of *D. malayana* with the present species.

It is presumable that this pigmy form is a species transported by some agencies into these geologically younger islands from some neighbouring older ones such as Philippine, Melanesia and Malay Archipelago. If it is really so, *D. malayana* and the present species are more closely related from the viewpoints both morphological and geographical.

Subfam. Megascolecinae

Genus *Pheretima* KINBERG

Pheretima sp.

Locality and material: Lelo, Kusaie, Caroline Isls., Dec. 10, 1937.
A single juvenile specimen.

Both externally and internally, the genital organs and their markings are not yet developed. Thus the identification of the species was not possible. But, one pair of simple, finger-shaped intestinal caeca was seen to be fairly well-developed. Judging from the presence of this organ together with that of some other features, it is clear that the present specimen is of the species belonging to the MICHAELSEN's subgenus *Pheretima*.

LITERATURE

- 1) MICHAELSEN, W. (1900). Das Tierreich. Oligochaeta. Berlin.
- 2) — (1910). Oligochaeten von verschiedenen Gebieten. Mitt. naturh. Mus. Hamburg, 27.
- 3) — (1910). Die Oligochätenfauna der vorderindisch-ceylonischen Region. Abh. Ver. Hamburg, XIX.
- 4) — (1913). Die Oligochäten von Neu-Caledonien und den benachbarten Inselgruppen. F. Sarasin & J. Roux, Nova Caledonia, Zool., I, 5.
- 5) — (1913). Die Oligochaeten Columbias. Mem. Soc. neu. Sci. nat., V.
- 6) PICKFORD, G. E. (1932). Oligochaeta, Part II: Earthworms. Discovery Reports, IV.
- 7) — (1938). Earthworms in Yucatan Caves. Carnegie Inst. Washington Pub., 491.
- 8) STEPHENSON, J. (1923). Fauna of British India, Oligochaeta. London.
- 9) — (1924). Oligochaeta of the Siju Cave, Garo Hills, Assam. Rec. Ind. Mus., XXVI, 1.
- 10) — (1931). Description of Indian Oligochaeta. H. ind., XXXIII, 2.
- 11) UDE, H. (1905). Terricole Oligochäten von den Inseln der Südsee und verschiedenen andern Gebieten der Erde. Z. wiss. Zool., LXXXIII.

BEOBACHTUNGEN EINIGER THIOTROPHER SEEN JAPANS MIT BESONDERER BERÜCKSICHTIGUNG DER SCHWEFELBAKTERIEN. II

VON

TADAO JIMBO

Biologisches Institut der Kaiserlichen Tohoku Universität in Sendai

Mit 1 Textfigur

Eingegangen am 25. November 1939

Ich habe schon früher eine zusammenfassende Darstellung unserer derzeitigen Kenntnisse über Schwefelwasserstoff-Seen auf Grund der bisherigen Forschungen gegeben. Darin sind drei von den japanischen Brackwasserseen, die verschiedene Typen vertreten und an denen ich bereits vorläufige Beobachtungen gemacht habe, eingehend besprochen. (Vgl. JIMBO 1938.)

Neuerdings hatte ich Gelegenheit, unseren einzigen thiotrophen Gebirgssee Wakuike bezüglich der Schwefelbakterien zu untersuchen. Hier mochte ich die dabei erhaltenen Ergebnisse mitteilen. Die Kosten der vorliegenden Arbeit wurden zum Teil durch ein Stipendium von der Japanischen Gesellschaft zur Förderung der Wissenschaften gedeckt. Dafür bin ich der genannten Gesellschaft zu aufrichtigem Danke verpflichtet. Ausserdem wurde ich bei der Durchführung der Beobachtungen von den Herren G. MINAMISAWA und K. MURATA in lebenswürdiger Weise unterstützt, wofür ihnen an dieser Stelle der verbindlichste Dank ausgesprochen sei.

Südwestlich der Stadt Nagano, inmitten eines bergigen Geländes, befindet sich der Berg Iwakura (761 m ü. d. M.), welcher dadurch bekannt ist, dass besonders sein Südwesthang geologisch ungemein labil ist und so der an seinem Fuss laufende Sai-Fluss (100 m ü. d. M.) mehrmals bei Erdbeben durch herabfallende Erdschollen gefüllt worden ist. Ein grosses Erdbeben rief im Jahre 1817 eine von der verlängerten Sperrung des Stromes bewirkte Katastrophe hervor, und schuf in halber Höhe dieses Südwesthanges des Berges Iwakura bei 580 m ü. d. M. den kleinen See Wakuike, von dem im vorliegenden Aufsatz die Rede ist.

Wie bei vielen sonstigen Seen in Japan, ist die allererste Lotung im Wakuike von TANAKA (1926) durchgeführt worden. Er hat dabei vor

allem entdeckt, dass das Seewasser ~~sehr~~ reich an Salzen, insbesondere an Gips, ist, und dass das Tiefenwasser Schwefelwasserstoff enthält. Darauf sind die Eigentümlichkeiten dieses Sees durch gründliche Untersuchungen von YOSHIMURA (1936) und UENO (1936) fast allseitig klargelegt worden. In Wirklichkeit ist dieser, wie sein Name ausdrückt (Waku-Ike quellender Weiher), mit unterseeischem Quellwasser gespeiste See zu den Salzseen zu rechnen.



Sommerbild von Waku-Ike, von Osten gesehen, im Hintergrund sich jenseits des Tales des San-Flusses erhebende Gebirge, worüber man bei schönem Wetter die Gipfel der „japanischen Alpen“ schauen kann.

Leider wissen wir über die Schwefelbakterien dieses Schwefelwasserstoff-Sees und die Lage im Winter nichts. Diese Lücken auszufüllen, ist das Ziel der vorliegenden Arbeit.

Bei diesem kaum 2,3 ha¹ messenden, rundlichen See, fehlt oberirdischer Abfluss und auch die zwei Zuflüsse sind recht unbedeutend. Indem das Seewasser während des Sommers zur Bewässerung benutzt wird, sinkt der Wasserstand, der im Spätfrühling das Maximum erreicht, im Laufe dieses Zeitraumes sehr stark herab, so dass die Wassertiefe beim Niedrigwasserstand um ca. 4 m flacher als die Maximaltiefe von 10,8 m ist.

Ich habe im Jahre 1939 zweimal den See besucht, erst im Februar und dann im August. Im Februar war das Wasser unter Eis bei weitem

¹ Darüber habe ich in der früheren Schrift eine irrtümliche Angabe gemacht.

klarer als im Hochsommer. So betrug die Sichttiefe im Februar 3 m, im August 0,7 m. Übrigens ist alles, was ich jedesmal an einer profunden Stelle bestimmt habe, in nachstehender Tabelle zusammengestellt¹⁾. Bemerkenswert ist, dass sich Schwefelwasserstoff nur im Sommer anhäuft.

Datum	26. II.			6. VIII.		
	11 ^h : über 8,3 m Tiefe			16 ^h : über 5,7 m Tiefe		
Tiefe m	Temperatur °C	H ₂ S	O ₂ cm/l	Temperatur °C	H ₂ S mg/l	Rötliche Färbung durch Chromatien
0	0,4	0	8,22	29,3	0	—
1	3,4		2,05	25,2	0	—
1,5				20,1		—
2	3,5	0	0,88	13,3	Spur	##
2,5				9,1		###
3	3,5		0,74	8,0	6	++
4	3,5			7,5	12	+
5	3,5			7,1	15	—
6	3,5	0	0,73			
7	3,5	0	0,75			
7,5	3,5	0	0,67			
8	3,6					

Damit haben wir nunmehr das Vorhandensein der Vollzirkulation im Herbst bei diesem See kennen gelernt. Ich habe im Sommer ein Massenschwärmen von *Chromatium Weissei*, einer schwefelführenden Purpurbakterienart, und *Chloronium mirabile*, einer äusserst eigenartigen, beweglichen Grünbakterie²⁾, hauptsächlich an der oberen Grenze der schwefelwasserstoffhaltigen unteren Wasserschicht gefunden. Das Wasser der betreffenden Tiefe zeigt deutliche pfirsichblütrote Farbe, die von den Chromatien hervorgerufen wird, während die auf den Grünbakterien beruhende grüne Nuance im Hintergrunde bleibt. Gleichwohl sind die letzteren ebenso zahlreich wie die ersteren, und ferner werden diese die Chromatien begleitenden Grünbakterien niemals ausserhalb der schwefelwasserstoffhaltigen Wasserschicht gefunden. Diese Tatsache liefert einen Beweis dafür, dass sich diese Grünbakterienart auf ganz dieselbe Weise wie die Chromatien verhält, insofern als sie mindestens in ihren normalen

¹⁾Das Temperaturprofil und die maximale Schwefelwasserstoffkonzentration im Sommer stehen im grossen und ganzen im Einklang mit YOSHIMURAS Befund, worin die letztere 18,9 mg/l beträgt.

²⁾Hierzu ist auf die Schilderungen von BUDER (1913) und von GEITLER und PASCHER (1925) zu verweisen.

Standorten den Schwefelwasserstoff nebst dem Lichte verlangt.

In der Literatur liegen meines Wissens noch keine Angaben über das Vorkommen derartiger Grünbakterienplatten im See vor, obgleich sie bezüglich der Achromatien bisher nicht fehlen. Was die chemosynthetischen farblosen Schwefelbakterien anbetrifft, so liegen die Verhältnisse einigermaßen anders als bei den Purpur- und Grünbakterien. Bei ihnen, die anstatt des Lichtes Sauerstoff fordern, ist ihre Ansammlung an der Grenzschicht auf die Bedürfnisse nach Sauerstoff einerseits und Schwefelwasserstoff andererseits zurückzuführen. Allerdings ist die photosynthetische Natur der Grünbakterien kaum zu bezweifeln. Aber man kann darüber im Zweifel sein, ob diese Grünbakterie photo-heterotroph wäre und wegen ihrer blossen anaeroben Beschaffenheit aus der schwefelwasserstoffhaltigen Wasserschicht nicht austreten könnte. Wenn auch der See tatsächlich sehr eutroph ist¹⁾, so scheint es mir doch unwahrscheinlich, dass eine so üppige Entwicklung dieser Bakterie durch wirklich darin gelöste organische Substanzen unterstützt würde. Hier ist indessen zu berücksichtigen, dass es sich bei *Chloronium mirabile* nicht um einzelne Bakterienzellen, sondern um eine Vereinigung eines zentral gelegenen, begeißelten, farblosen Stäbchens mit mehreren umliegenden, geißellosen, grünen Stäbchen handelt. Besonders beachtenswert ist die Tatsache, dass die ganze Vereinigung nicht den grünen Komponenten selbst, sondern dem farblosen zentralen Stäbchen ihre Bewegung verdankt. Infolge der Schwierigkeit der Kultur sind wir heute nicht in der Lage, die Natur des zentralen Stäbchens aufzuklären und das Wesen seiner taktischen Bewegungen kennen zu lernen. Jedoch ist anzunehmen, dass jedenfalls das Verhalten des zentralen Stäbchens mit den Ansprüchen der grünen Stäbchen nicht im Widerspruch stehen darf.

Der schwarze Tiefenschlamm riecht nach Schwefelwasserstoff ebenfalls im Winter, zur Zeit wo dieses Gas im Seewasser nicht vorhanden ist. Bei wochenlangem Aufbewahren davon mit etwas Bodenwasser zusammen in einem in eine beleuchtete warme Kammer gestellten Fläschchen, habe ich Massenentwicklung daraus von *Chromatium Weissei* und *Chlorobium limicola*, einer gewöhnlichsten grünen Stäbchenbakterie, wahrgenommen, was auch im Sommer bei dem *Chlorobium* vorgekommen ist. Daraus geht hervor, dass die Chromatien u. a. im Schlamm überwintern. Das unbewegliche *Chlorobium* ist natürlich nicht imstande, sich phototaktisch an der Grenzschicht anzusammeln.

¹⁾ Vgl. hierüber YOSHIMURA (1936) und UÉNO (1936).

ZUSAMMENFASSUNG

1. Ein Massenschwärmen am Metalimnion von *Chromatium Weissei* und grünem *Chloronium mirabile* wurde an Wakuike, einem mitteljapanischen Salzsee, ermittelt, wo sich Schwefelwasserstoff nur im Sommer anhäuft.

2. Diese Tatsache spricht dafür, dass diese Grünbakterien, ebenso wie die Chromatien, Schwefelwasserstoff und Licht verlangen.

3. Ferner wurde unbewegliches, grünes *Chlorobium limicola* im Tiefenschlamm gefunden.

LITERATUR

- BUDER, J. (1913): *Chloronium mirabile*. Ber. d. Deutsch. Bot. Gesell. **31**, 80.
GEITLER, L. und A. PASCHER (1925): Cyanochloridinae. PASCHER, Süßwasserflora. Heft 12, 451. (Vgl. p. 460.)
JIMHO, T. (1938): Beobachtungen einiger thiotropher Seen Japans mit besonderer Berücksichtigung der Schwefelbakterien. Sci. Rep. Tōhoku Imp. Univ. 4. Ser. **13**, 259.
TANAKA, A. (1926): Untersuchungen des Nojiri-Sees. (Japanisch.)
UÉNO, M. (1936): Productivity of an extremely eutrophic lake in middle Japan. Proc. Imp. Acad. Tōkyō. **12**, 248.
YOSHIMURA, S. (1936): Limnology of Lake Wakuike, Nagano Prefecture, Japan. Proc. Imp. Acad. Tōkyō **12**, 245.

ON THE MULTIPARTITE CHROMOSOME-RING IN *CEPHALOTAXUS DRUPACEA* SIEB. ET ZUCC.

By

YOSINORI SUGIHARA

Biological Institute, Tôhoku Imperial University, Sendai

(With Plates I-II)

(Received November 27, 1939)

INTRODUCTION

Concerning the chromosome number of *Cephalotaxus drupacea*, LAWSON ('07) reported that it may be ten in haploid and twenty in diploid generation, but ISHIKAWA ('16) stated, by the observation of the meiosis of the pollen mother-cell, the haploid chromosome number of this plant to be twelve. Recently SAX and SAX ('33) described in *Cephalotaxus Fortunei* that the chromosome number of this plant is twelve in the mitosis of the endosperm.

The writer's occasional observations to determine the chromosome number of *C. drupacea* had revealed that in certain individuals of this plant a multipartite chromosome-ring appears regularly in the first division of the meiosis. So last spring a detailed investigation was made on this subject. The results obtained will be described in the present paper.

Since the publication of CLELAND's research concerning the multipartite chromosome-ring of *Oenothera franciscana* similar cases have been reported in a number of flowering plants. But in Gymnosperms, as far as the writer is aware, as a single example, MATSUURA and SUTÔ ('35) reported a case in *Taxus cuspidata*. In the meiosis of the male plants of this species, ten bivalents and an association of four chromosomes were observed.

In this occasion the writer wishes to express his cordial thanks to Professor Dr. M. TAHARA for his helpful suggestions and criticisms during the course of this investigation. Thanks are also due to Mr. T. TERASAKI who collected the material from Tokyo.

MATERIAL AND METHOD

For the present investigation thirty four male individuals were used. The localities of the individuals are as follow:

Individual-number	Locality
1	In the grounds of the Biological Institute, Tôhoku Imperial University, Sendai
2 21	Tenshudai, Sendai.
22, 23, 25 32*	Rifu (near Sendai), Miyagi Prefecture
33-34	Miyaginohara, Sendai.
35	The Koiskawa Botanic Garden of Tokyo Imperial University.

For the observation of the meiosis in the pollen mother-cells, the smear preparations were used. For fixative, TAYLOR's solution was used, but chromic acid was reduced to about half quantity and maltose was not added at all. For staining NEWTON's gentian violet iodine was used. A slight pressure of the finger on the cover glass was very effective for the scattering of chromosomes. This enabled the writer to distinguish each chromosome with great ease.

OBSERVATION AND CONCLUSION

In the first place the haploid chromosome number of this plant was estimated to be twelve by observation of the metaphase of the first division of the meiosis in pollen mother-cells (Pl. I, fig. 1; Pl. II, fig. 6). The writer's attention was next directed to the constant associations of four or six chromosomes in certain individuals. Thorough studies of the thirty four individuals have shown that as for the association of chromosomes five different types can be distinguished. For the sake of brevity they are shown as follow: 12_{II} , $10_{II} + 1_{(4)}$, $8_{II} + 2_{(4)}$, $7_{II} + 1_{(4)} + 1_{(6)}$ and $5_{II} + 2_{(4)} + 1_{(6)}$ ** (Pl. I, figs. 1-5). Nine out of the thirty four individuals constantly formed only twelve bivalents in the meiosis (Table I; Pl. I, fig. 1; Pl. II, fig. 6) and in the remaining twenty five individuals, the chromosomal associations of four or six chromosomes were found (Table

Table I. Individuals of the Type, 12_{II}

Individual-number	Number of examined nuclear plates	Frequency of 12_{II}	Frequency of other associations
5	131	131	0
21	106	106	0
23	114	112	2 (11_{II} , $2_{(4)}$)
26	119	117	2 (11_{II} , $2_{(4)}$)
29	103	103	0
32	25	25	0
33	20	20	0
34	20	20	0
35	8	8	0

* The plant, No. 24 was very poor in growth and could not be used for this study.

** (4) or (6) denotes a ring or chain of 4 or 6 chromosomes.

Table II. Individuals of the Type, $10_{II} + 1_{(4)}$

Individual number	Number of examined nuclear plates	Frequency of $10_{II} + 1_{(4)}$	Frequency of other associations
12	101	101	0
13	35	35	0
14	25	25	0
20	117	115	2 { 9_{II} , $1_{(4)}$, $2_{(1)}$ }
22	3	3	0 { 9_{II} (ring), 3_{II} (rod) }
25	18	18	0
27	25	25	0
28	15	15	0
30	3	3	0
31	14	14	0

Table III. Individuals of the Type, $8_{II} + 2_{(4)}$

Individual number	Number of examined nuclear plates	Frequency of $8_{II} + 2_{(4)}$	Frequency of other associations
1	18	18	0
2	11	11	0
3	21	20	1 { 8_{II} , 1 chain of 4, 2_{II} (rod) }
11	25	25	0

Table IV. Individuals of the Type, $7_{II} + 1_{(4)} + 1_{(6)}$

Individual number	Number of examined nuclear plates	Frequency of $7_{II} + 1_{(4)} + 1_{(6)}$	Frequency of other associations
4	11	11	0
7	36	35	1 { 7_{II} , 6 rings, 1 rod, $1_{(4)}$, 2 chains of 3 }
9	9	7	2 { 8_{II} , 6 rings, 2 rods, $1_{(4)}$, 1 chain of 4 }
10	31	30	2 { 7_{II} (ring), $1_{(4)}$, 1 chain of 5, $1_{(1)}$ }
15	28	28	1 { 8_{II} (6 rings, 2 rods), $1_{(4)}$, 1 chain of 4 }
16	30	30	0
17	20	20	0
18	27	25	0 { 7_{II} (ring), $1_{(4)}$, 2 chains of 3 }
19	6	6	2 { 9_{II} (6 rings, 3 rods, $1_{(6)}$) }

Table V. Individuals of the Type, $5_{II} + 2_{(4)} + 1_{(6)}$

Individual number	Number of examined nuclear plates	Frequency of $5_{II} + 2_{(4)} + 1_{(6)}$	Frequency of other associations
6	24	24	0
8	25	25	0

II-V; Pl. I, figs. 2-5; Pl. II, figs. 7-10).

The type of the association of the chromosomes was, in most cases, constant in a definite individual, with only a very few exceptions (Table I,

No. 23, 26; Table II, No. 20; Table III, No. 3 and Table IV, No. 7, 9, 10, 18) and in none of these exceptional instances was it the case, that the exceptional association could be considered as having been caused by the alteration of the chromosomal constitution. On the contrary, these exceptional cases appear, in all probability, to have arisen from an occasional failure of the association or from an accidental breakage of a multipartite chromosome-ring. For example in one individual among the group of the constitution, $10_{II}+1_{IV}$, one nuclear plate was met with showing only twelve bivalents (Table II). Nine of these twelve bivalents were ring-shaped, and the remaining three were rod-shaped. Probably two out of these three had come into existence by the breakage of a tetrapartite chromosome-ring. The bivalents which are regarded as having appeared as the result of the breakage of a multipartite chromosome-ring, are, without exception, rod-shaped. It is important, however, that all rod-shaped bivalents should not be regarded as the derivatives of the multipartite chromosome-ring, because, as is shown in Table VI, the

Table VI. Frequency of the appearance of rod-shaped bivalents

Num- ber of rod- shaped bivalents in a nuclear plate	Individual- number	Type, 12_{II}										Type, $10_{II}+1_{IV}$									
		5	21	23	26	29	32	33	34	35		12	13	14	20	22	25	27	28	30	31
0		98	70	94	68	66	16	13	9	8		66	26	14	91	3	10	17	13	3	11
1		27	25	19	36	26	6	7	7	0		33	9	10	25	0	6	6	2	0	2
2		6	11	1	14	11	2	0	4	0		2	0	1	1	0	1	2	0	0	1
3		0	0	0	1	0	1	0	0	0		0	0	0	0	0	1	0	0	0	0

Num- ber of rod- shaped bivalents in a nuclear plate	Individual- number	Type, $8_{II}+2_{IV}$					Type, $7_{II}+1_{IV}+1_{VI}$										Type, $5_{II}+2_{IV}+1_{VI}$	
		1	2	3	11	4	7	9	10	15	16	17	18	19			6	8
0		16	5	12	23	9	25	5	25	16	26	17	21	6			19	21
1		2	6	7	2	1	10	2	5	11	4	3	5	0			5	4
2		0	0	2	0	1	1	2	1	1	0	0	1	0			0	0
3		0	0	0	0	0	0	0	0	0	0	0	0	0			0	0

normal bivalents sometimes take a rod-shape. So the estimation of the frequency of the appearance of the rod-shaped bivalents seems necessary for the determination of the chromosomal constitution. But in reality the frequency of their appearance was very low in all the individuals of

each type (Table VI). Bivalents with three or more chiasmata were never found. In rare exceptional cases bivalents have appeared as univalents. The association of four or six chromosomes is found, in its configuration, generally in a ring, and rarely in a chain (Table VII).

Table VII. Percentage, in which the association, (4) or (6) occurs in chain configuration

Type	% in chain configuration
$10_{II} + 1_{(4)}$	1.3
$8_{II} + 2_{(4)}$	3.5
$7_{II} + 1_{(4)} + 1_{(6)}$	12.4
$5_{II} + 2_{(4)} + 1_{(6)}$	17.9

In conclusion, in *Cephalotaxus drupacea* there are among the different individuals different chromosomal constitutions which are characteristic of each individual. Such a heterogeneity found among the individuals belonging to the same species has arisen perhaps by segmental interchange of the chromosomes. Similar cases have been already reported in *Datura stramonium* (BLAKESLEE '28, BLAKESLEE, BERGNER & AVERY '37 etc.), *Campanula persicifolia* (GAIRDNER & DARLINGTON '31, DARLINGTON & GAIRDNER '37), *Zea Mays* (MCCLINTOCK '30 etc.), *Pisum sativum* (HÅKANSSON '31, SANSOME '32 etc.), *Lilium Hansonii* (HAGA '38) and others.

It is remarkable that in *Cephalotaxus drupacea* the plants of the five different chromosomal constitutions are found in close proximity in a very small restricted area. Whether outer morphological differences in conformity with the internal chromosomal differences are or are not to be observed can not be decided at present. It is highly possible that further research of this plant will discover the existence of still other types of the chromosomal constitution, for example $9_{II} + 1_{(6)}$.

SUMMARY

In the metaphase of the first division of the pollen mother-cells of certain individuals of *Cephalotaxus drupacea*, special chromosomal associations were found. In this respect five types were distinguished in this plant, namely 12_{II} , $10_{II} + 1_{(4)}$, $8_{II} + 2_{(4)}$, $7_{II} + 1_{(4)} + 1_{(6)}$ and $5_{II} + 2_{(4)} + 1_{(6)}$. The type of association is constant in each individual. The multipartite chromosome-ring or chain should be regarded as having been caused by segmental interchange among the chromosomes.

LITERATURE

- BLAKESLEE, A. F., 1928. Genetics of *Datura*. Zeitsch. ind. Abst. u. Vererb., Supplbd. 1: 117-130.
- BLAKESLEE, A. F., A. D. BERGNER & A. G. AVERY, 1937. Geographical Distribution of Chromosomal Prime Types in *Datura stramonium*. Cytologia, Fujii Jub. Vol.: 1070-1093.
- CLELAND, R. E., 1922. Reduction Division in the Pollen Mother Cells of *Oenothera franciscana*. Amer. Jour. Bot., 9: 391-413.
- DARLINGTON, C. D. & A. E. GAIRDNER, 1937. The Variation System in *Campanula persicifolia*. Jour. Genet., 35: 97-128.
- GAIRDNER, A. E. & C. D. DARLINGTON, 1931. Ring Formation in Diploid and Polyploid *Campanula persicifolia*. Genetica, 13: 113-150.
- HAGA, T., 1938. A Reciprocal Translocation in *Lilium Hansonii* LEICHT. (A Prelim. Note) Japanese with English Résumé. Jap. Jour. Genet., 14: 296-299.
- HÂKANSSON, A., 1931. Über Chromosomenverkettung in *Pisum*. Hereditas, 15: 2-61.
- ISHIKAWA, M., 1916. A List of the Number of Chromosomes. Bot. Mag. (Tokyo), 30: 404-448.
- LAWSON, A. A., 1907. The Gametophytes, Fertilization and Embryo of *Cephalotaxus drupacea*. Ann. Bot., 21: 1-23.
- MATSUURA, H. & T. SUTÔ, 1935. Contributions to the Indigram Study in Phaenerogamous Plants. I. Jour. Fac. Sci. Hokkaidô Imp. Univ., Ser. V, 5: 33-75.
- MCCLEINTOCK, B., 1930. A Cytological Demonstration of the Location of an Interchange between the Non-homologous Chromosomes of *Zea Mays*. Proc. Nat. Acad. Sci., 16: 791-796.
- SANSOME, E. R., 1932. Segmental Interchange in *Pisum sativum*. Cytologia, 3: 200-219.
- SAX, K. & H. J. SAX, 1933. Chromosome Number and Morphology in the Conifers. Jour. Arnold Arb., 14: 356-375.

EXPLANATION OF PLATES

PLATE I

Five types of chromosomal association in meiotic metaphase. $\times 2070$.

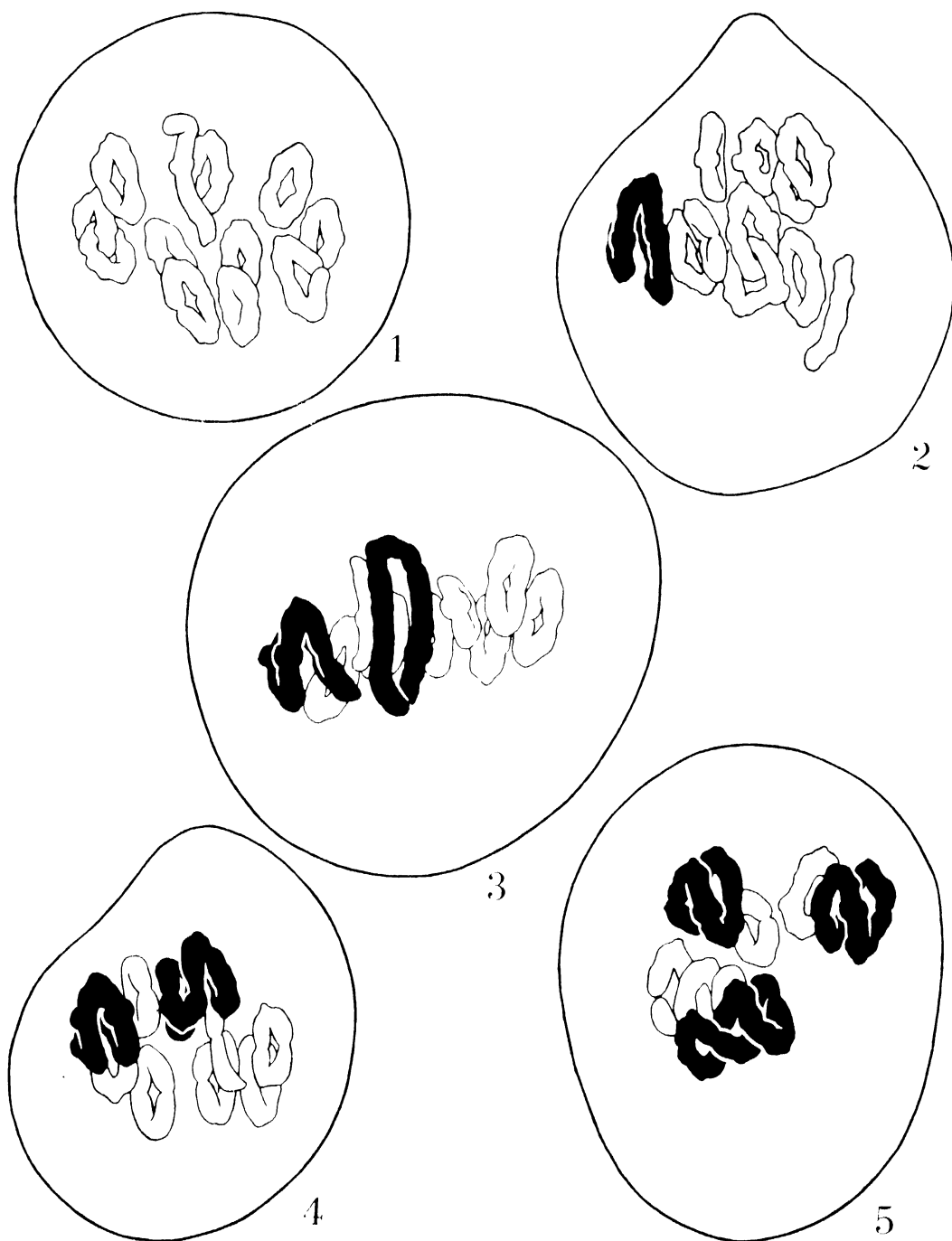
- Fig. 1. Type 12_{II} (Individual-number 5).
- Fig. 2. Type $10_{II}+1_{(4)}$ (Individual-number 12).
- Fig. 3. Type $8_{II}+2_{(4)}$ (Individual-number 1).
- Fig. 4. Type $7_{II}+1_{(4)}+1_{(6)}$ (Individual-number 4).
- Fig. 5. Type $5_{II}+2_{(4)}+1_{(6)}$ (Individual-number 6).

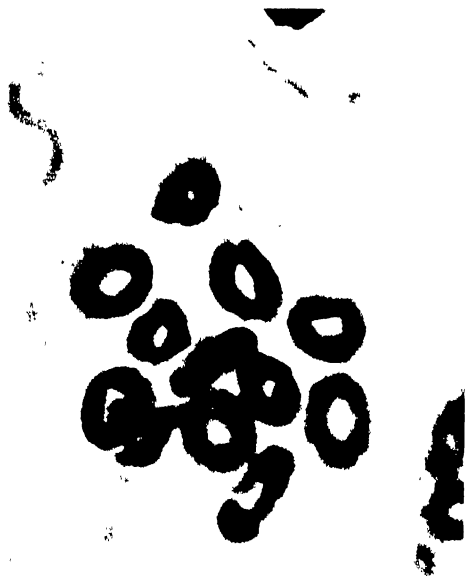
PLATE II

Microphotographs of the three different types of chromosomal association.

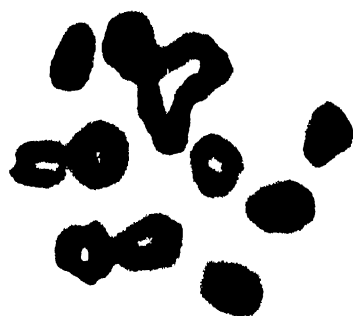
Figs. 6-8 $\times 2220$. Figs. 9-10 $\times 4470$.

- Fig. 6. Type 12_{II} (Individual-number 5).
- Fig. 7. Type $10_{II}+1_{(4)}$ (Individual-number 20).
- Fig. 8. Type $5_{II}+2_{(4)}+1_{(6)}$ (Individual-number 6).
- Fig. 9. Ring of four chromosomes (Individual-number 6).
- Fig. 10. Ring of six chromosomes (Individual-number 6).





6



7



8



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10

THE GAMETOPHYTES, FERTILIZATION AND PROEMBRYO OF *SCIADOPITYS VERTICILLATA*

By

MASATO TAHARA

Biological Institute Tōhoku Imperial University, Sendai

With Plate III and 10 text-figures

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In the year 1937 the present writer published an account concerning the gametophytes and embryo of *Sciadopitys verticillata*, a conifer endemic to Japan. To make further studies of this plant materials were collected in the succeeding years. The observations based on these materials will be presented in the following pages. As in the preceding research, the materials were collected mainly from a large tree standing in the grounds of the Kameoka-Hachiman, a shrine in the suburbs of Sendai.

THE MALE GAMETOPHYTE

The meiotic division in the pollen-mother-cells occurs early in March. For the examination of this division the smear method was employed. The fixation was made by TAYLOR's solution and the staining by NEWTON's gentian-violet iodine. The division proceeds normally. Ten ring-shaped gemini were easily counted in the metaphase of the first division of the meiosis (Fig. 1).

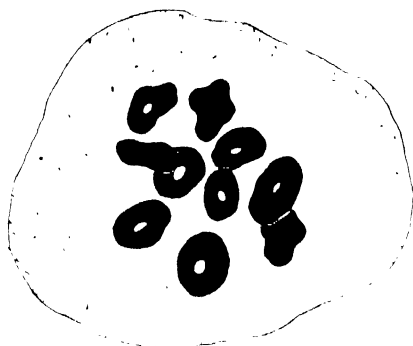


Fig. 1. 1st division of the meiosis in a pollen mother cell. $\times 1700$.

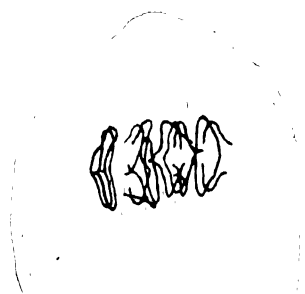


Fig. 2. Mitosis at the upper extremity of the body cell. $\times 850$.



Fig. 3, a. The body cell is divided into two unequal cells. $\times 400$.

cell is found near the stalk nucleus and the pollen tube nucleus. In most species of *Taxodiaceae* and *Cupressaceae* the body-cell is divided into two equal cells to form two functional sperm cells. But in the plant under observation the nuclear division occurs either at the upper or at the lower extremity of the cells. So the resulting two cells are very unequal in size (Fig. 3, a and b). The smaller one degenerates in the meantime and is not able to fertilize the egg nucleus. Later on, this cell is often seen at the side of the large functioning sperm cell (Fig. 4).

Pollination takes place later in April. Pollen grains at this time contain two nuclei, a pollen tube nucleus and a generative nucleus. There is no trace of vestigial prothallial cells. Later in June the generative nucleus divides, giving rise to the body cell nucleus and the stalk cell nucleus; meanwhile the latter moves towards the apex of the pollen tube, leaving the body cell in the original position.

The male gametophyte passes the winter in this condition. And just before the time of fertilization which occurs at the beginning of June of the next year, the body cell divides to form the sperm cells (Fig. 2). At this time the body



Fig. 3, b. The same. $\times 300$.

Concerning the sperm cell of this plant LAWSON describes as follows:

"...there was no cell membrane separating these two male nuclei from one another. The two structures were found lying quite freely in the cytoplasm of the body-cell and one of them is slightly but distinctly larger than the other."

THE FEMALE GAMETOPHYTE

At the end of May a single functional megaspore-mother-cell is differentiated out of the surrounding sporogenous tissue. The tetrads resulting from the meiotic division are represented, as LAWSON (1910) remarks, by three cells in the axial line, the middle one containing two free nuclei (Figs. 5, a, b). The basal megaspore now enlarges and begins to form the endosperm. The process of endosperm formation in this plant is unique. At first, as usual, a thin protoplasmic layer containing free nuclei is formed and then the wall formation takes place (Fig. 5, c).

As is well known, in the Gymnosperms there are two types of wall-formation of the endosperm. In one type, at the final free nuclear division, walls are formed perpendicular to the megaspore membrane. Thus the cells have at first no wall on the side towards the centre of the megaspore cavity. Afterwards the periclinal walls are formed, but the innermost sides of the innermost cells remain open. The method continues, until the central cavity is entirely closed.

In the other type, the first walls, perpendicular to the megaspore membrane, extend to the middle of the megaspore cavity. So the long tube-like cells radiating from the centre are seen in this case. Later these cells are divided into numerous cells by subsequent divisions.

The wall formation of the endosperm in *Sciadopitys* does not conform to either of these two types. The cells formed at the periphery next to the megaspore membrane are arranged with no regularity, that is they are not arranged in radial rows. Moreover it is remarkable that some of the cells contain one large or two or more small nuclei, and on the innermost side towards the central cavity a thin protoplasmic layer containing free nuclei is always seen during the whole course of the endo-

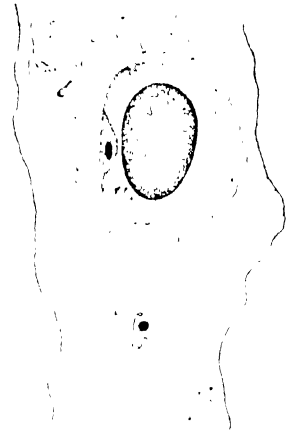


Fig. 4. The sperm cells. The smaller sperm cell is seen at the side of the larger functioning sperm cell. $\times 200$.

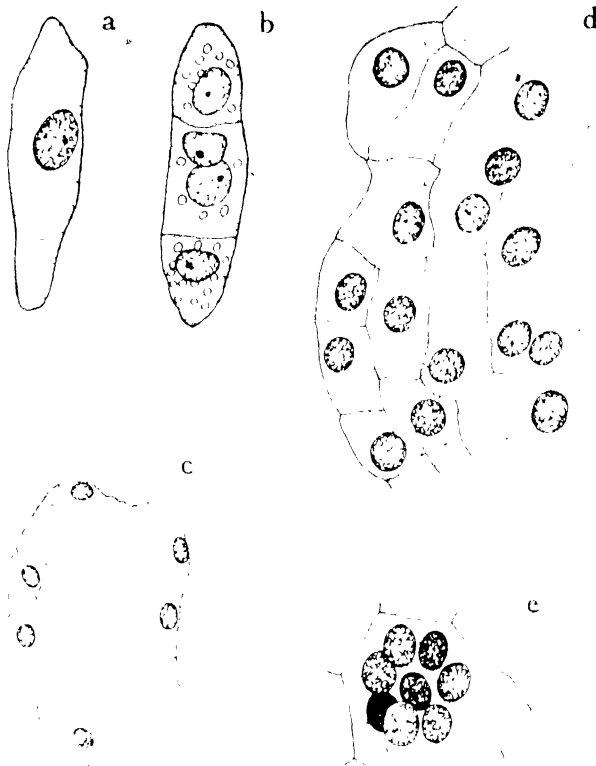


Fig. 5. Development of the female gametophyte. *a*, megaspore mother cell. *b*, axial row of megaspores, the middle one of which has two nuclei. *c*, early stage of endosperm formation. *d*, later stage of the same. *e*, an endosperm cell containing 8 nuclei. *a* and *b*, $\times 1100$. *c*, *d* and *e*, $\times 260$.

sperm formation; thus a definite layer of cells remaining open on the side towards the central cavity is not formed in this case (Fig. 5, *d*, *e*).

THE VENTRAL CANAL NUCLEUS AND FERTILIZATION

Shortly before fertilization, the nucleus of the central cell of the archegonium undergoes a division, which results in the formation of the egg nucleus and the ventral canal nucleus. Ten slender chromosomes which appear in this division can be counted without difficulty both in the side view and in the polar view. At the telophase of this division the nuclear membrane is usually formed only in the lower chromosome group (Fig. 6, *a*), i.e. the ventral canal nucleus is not formed at all. But in some exceptional cases the nuclear membrane is formed in both regions (Fig. 6, *b*). And the ventral canal nucleus remain intact for a

long time (Fig. 6, c). Sometimes 2, 4 or 8 supernumerary nuclei are seen in the middle or in the lower part of the archegonium, before the entrance of the sperm nucleus (Fig. 6, d). In the writer's opinion they may perhaps be the products of the ventral canal nucleus. In some cases one or two of these nuclei are seen almost in contact with the

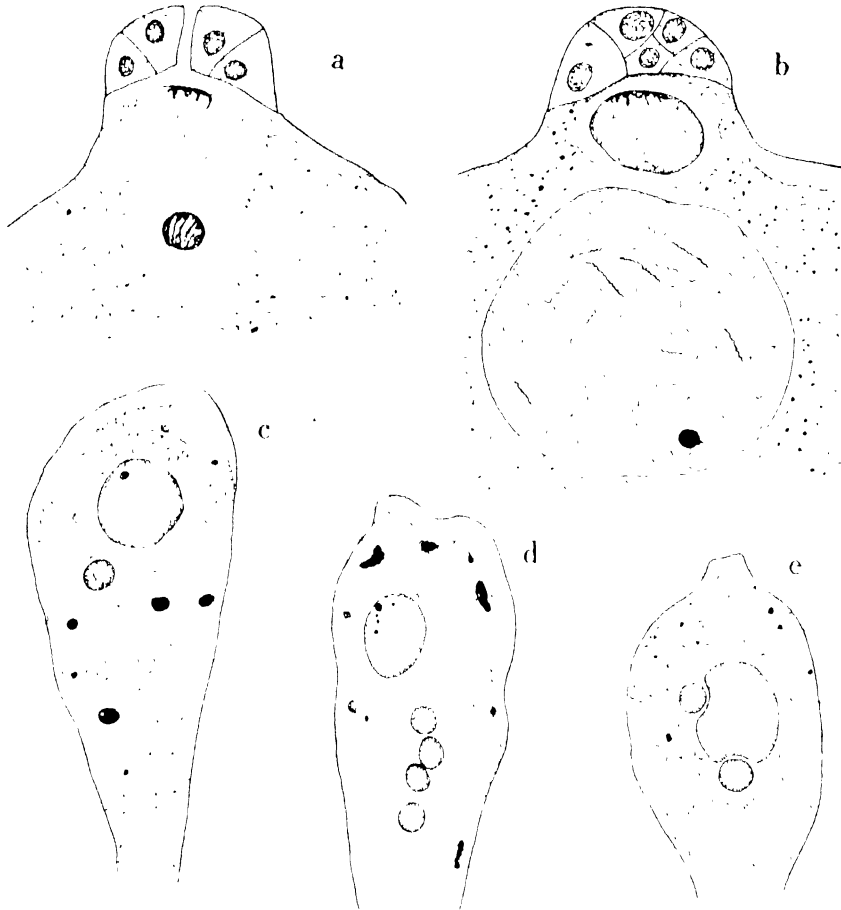


Fig. 6. *a*, telophase of the division in the central cell of the archegonium; the ventral canal nucleus is not organized. *b*, ventral canal nucleus and egg nucleus are both organized. *c*, egg nucleus and a ventral canal nucleus. *d*, egg nucleus and four small ventral canal nuclei. *e*, two small ventral canal nuclei almost in contact with the egg nucleus. *a*, *b*, $\times 240$. *c*–*e*, $\times 100$.

egg nucleus (Fig. 6, e). In one preparation, in the centre of the archegonium a fusion of two nuclei was observed (Fig. 7, b). This reminded the writer at first of the fertilization of the egg nucleus by a sperm

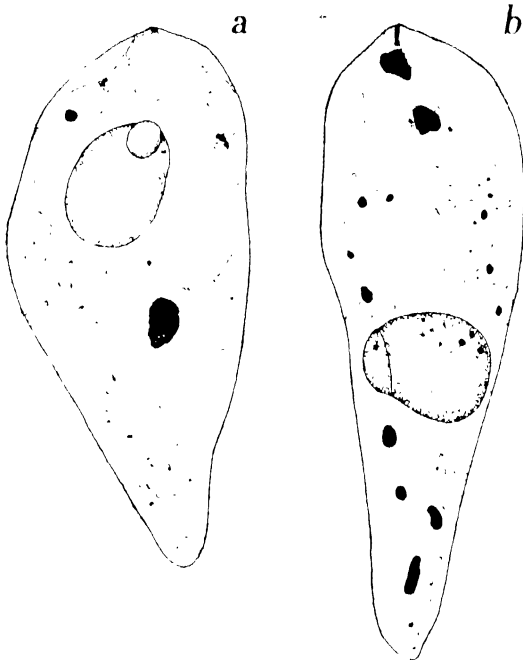


Fig. 7, *a*. Egg-nucleus fused with sperm nucleus.
b. Egg-nucleus fused with ventral canal nucleus. $\times 130$.

nucleus. But in this case the small nucleus was seen at side, not in the apex of the egg nucleus. So perhaps in this case, the egg nucleus is fused with the ventral canal nucleus. The figure which the writer considers as that of true fertilization is shown in Fig. 7, *a*. In this case the sperm nucleus which is very dense in its content is seen at the apex of the egg nucleus.

THE PROEMBRYO

Shortly after the fertilization the first mitosis for the formation of the proembryo occurs. Twenty chromosomes are counted in this mitosis (Fig. 8). The resulting two nuclei remain in the middle part of the archegonium and divide simultaneously to give rise to four nuclei which descend to the bottom of the archegonium. In this region four successive nuclear divisions take place one after another (Fig. 1-9, Pl. III). After the fifth mitosis we see 32 free nuclei, which become soon arranged in tiers, and wall formation begins. We can now distinguish three parts of the proembryo distinctly. The lowest part, consisting of about 13 cells is the principal part of the proembryo. The middle part consists of about 10 to 13 cells and are arranged in one layer. In these cells a wall is not formed on the side towards the



Fig. 8. Polar view of the metaphase of the first division after the fertilization $\times 1700$.

general cytoplasm of the archegonium, therefore the cells remain open on this side up to the sixth division. In the uppermost tiers no cell formation occurs; about 6 to 8 nuclei are found in free condition. Now the sixth nuclear division comes about. This division is not, in the strict sense, simultaneous, as in one part, either the upper or the lower, it takes place earlier than in the other. Moreover it seems to the writer probable that in this case some nuclei, especially the ones in the lowest part do not perform the mitosis at all. After this division we see nearly 60 nuclei composing



Fig. 9. The final stage of the proembryo development in four successive sections. $\times 175$.

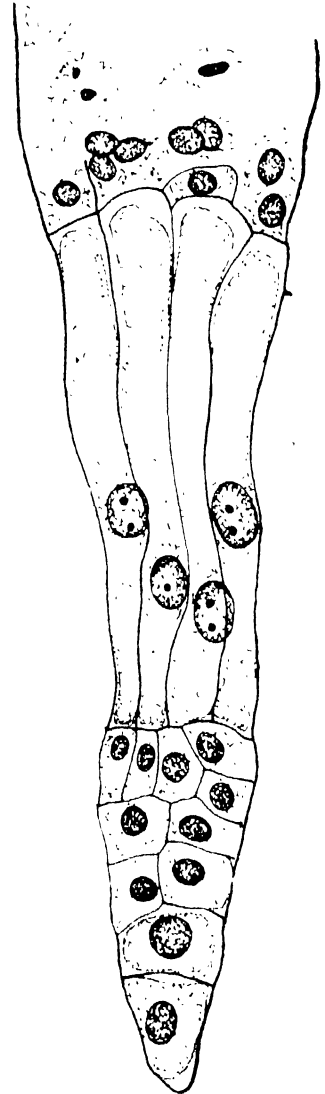


Fig. 10. The elongation of the prosuspensor. $\times 150$.

the final stage of the proembryo development. The middle part, one cell thick, is now surrounded completely by cell wall and later forms the prosuspensor. The number of the cells in this layer is the same as that in the previous stage. This is caused by the fact that in sixth division the nuclear spindles are directed, in this part of the proembryo, almost parallel to the long axis of the archegonium (Fig. 8, Pl. III). In the uppermost part we see free nuclei and incomplete cells which are open on the side towards the centre of the archegonium. Later a few of these open cells may be closed completely by the cell wall to form the so-called rosette cells and eventually the open cells disappear (Fig. 7, 8, Pl. III).

In conclusion the proembryo in the final stage consists of about 20 embryo-initials forming the lowest part of the embryo, 10 to 13 cells of the prosuspensor, 0 to 3 rosette cells and about 20 to 30 free nuclei.

In 1931 BUCHHOLZ gave an account relating to the later stages of the embryo formation in *Sciadopitys verticillata*. But his estimation as to the number of the cells in the different parts of the proembryo does not coincide exactly with the writer's,

He says: "The number of lower embryo initials is 12 to 18 or more; the number of cells in the prosuspensor is usually 7 to 9, the rosette cells 0 to 9 or more; and the number of free nuclei above the suspensor, if there are any, is unknown."

Among the conifers, in the first period of embryogeny, 32 or more free nuclei are formed in the Araucariaceae (EAMES, 1913; BURLINGAME, 1915). So in this respect *Sciadopitys* bears a great resemblance to this family. But in the structure of proembryo after the wall formation no similarity is observed in these two plants. In any case *Sciadopitys* is a plant which shows a pronounced complexity in the course of its early embryogeny.

SUMMARY

1. In *Sciadopitys verticillata* the body cell is divided into two unequal cells. The smaller one is formed in the upper or in the lower extremity of the body cell. This small cell later disintegrates and is found often on one side of the large functioning sperm cell.

2. In the endosperm formation, at first, a thin protoplasmic layer containing free nuclei is formed around the central cavity of the megaspore. In the meantime the cell-wall formation begins. The cells newly formed are, however, arranged with no regularity. The radial arrange-

ment of the cells, which is usual in most species of Gymnosperms is not seen in this plant. Moreover some of the endosperm-cells of this plant contain one large or two or more small nuclei already in the early stage of the endosperm development. The thin protoplasmic layer containing free nuclei is seen up to the final closure of the cavity.

3. In some exceptional cases the ventral canal nucleus remain intact for a long time. Sometimes 2, 4 or 8 supernumerary nuclei are found in the middle or in the lower part of the archegonium, before the entrance of the sperm nucleus. They are, in all probability, the products of the ventral canal nucleus.

4. In the course of the proembryo formation six simultaneous nuclear divisions take place one after another. The cell-wall formation begins after the fifth nuclear division, that is after the formation of 32 free nuclei and is completed after the sixth nuclear division. In the sixth nuclear division some nuclei remain in resting stage. So in the final stage of the proembryonal development we cannot definitely count 64 nuclei. The number of the cells in the lowest part, embryo initials, is about 20; the number of cells in the prosuspensor is 10 to 13 or more, the rosette cells 0 to 3, the free nuclei in the uppermost tier about 20 to 30.

LITERATURE CITED

- BUCHHOLZ, JOHN, T. (1931). The Suspensor of *Sciadopitys*. Bot. Gaz. **92**.
BURLINGAME, L. L. (1915). The Morphology of *Araucaria brasiliensis*. III. Bot. Gaz. **59**.
EAMES, A. J. (1913). The Morphology of *Agathis australis*. Ann. Bot. **27**.
LAWSON, A. A. (1910). The Gametophytes and Embryo of *Sciadopitys verticillata*. Ann. Bot. **24**.

EXPLANATION OF PLATE III

Development of proembryo in *Sciadopitys verticillata*. Magnification, $\times 200$.

Fig. 1. The 3rd mitosis after fertilization.

Fig. 2. The 8 nucleus stage.

Fig. 3. The 4th mitosis.

Fig. 4. The 16 nucleus stage.

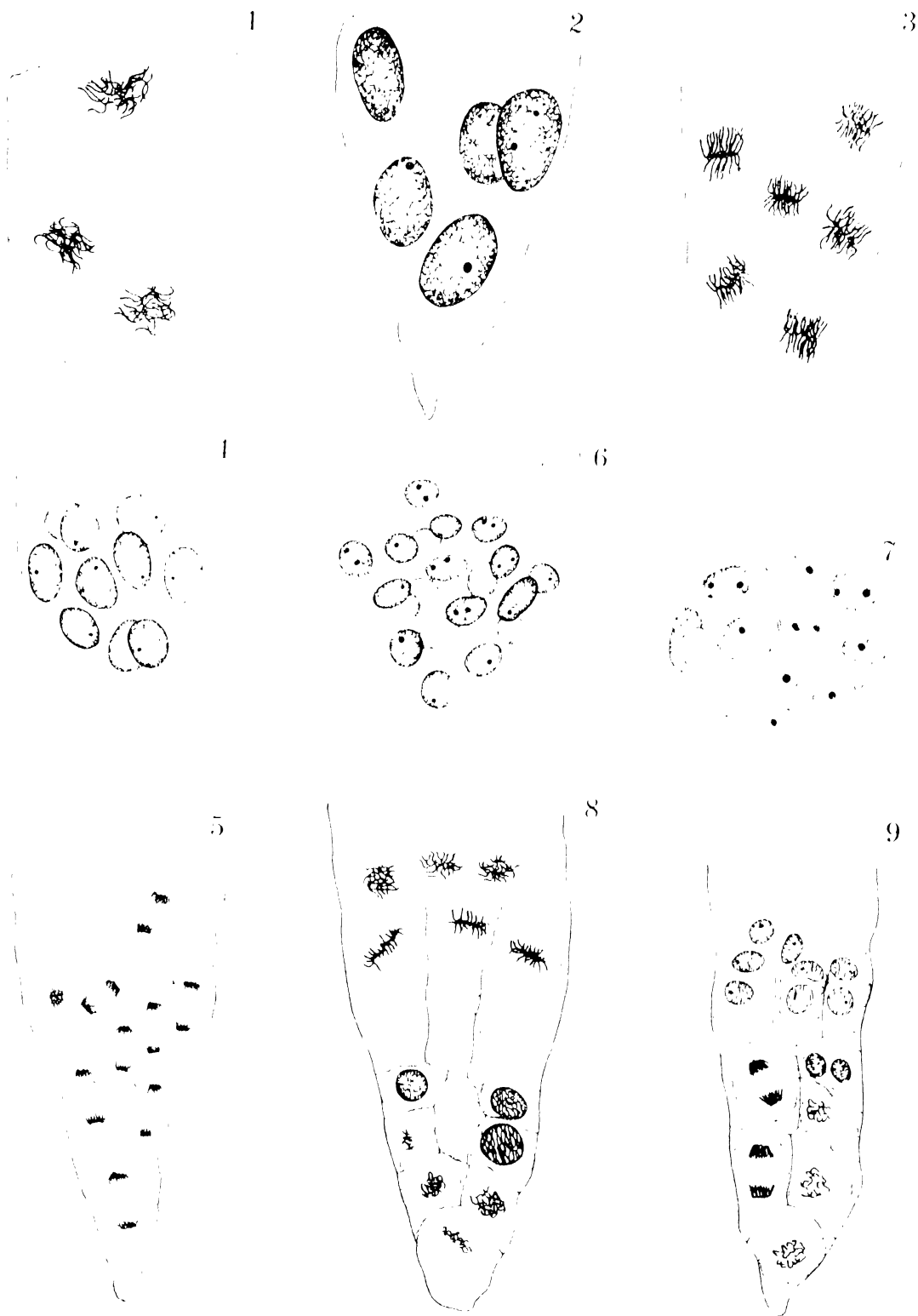
Fig. 5. The 5th mitosis.

Fig. 6. The 32 nucleus stage.

Fig. 7. Cross section of the middle part of the prosuspensor in 32 nucleus stage after the cell-wall formation.

Fig. 8. The 6th mitosis.

Fig. 9. The later stage of the 6th mitosis.



ON SOME CALCAREOUS SPONGES FROM JAPAN

By

SANJI HÔZAWA

Biological Institute, Tôhoku Imperial University, Sendai, Japan

(With Plates IV-V and 9 text-figures)

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In the present paper the writer wishes to report on some calcareous sponges obtained from Japan. The specimens upon which the following descriptions were based had been secured by him while on collecting tours in the summer of 1933 to Wagu in Miye Prefecture, to Wajima in Ishikawa Prefecture and to Oshima in Miyagi Prefecture. In undertaking the above tours he received financial support from the Japan Society for the Promotion of Scientific Research and to this society his sincerest thanks are due.

The following is the list of species dealt with in the present work.

Family Homocoelidae

1. *Leucosolenia clathrata* (CARTER)
2. *Leucosolenia ventosa*, n. sp.
3. *Leucosolenia reticulum* (O. SCHMIDT)
4. *Leucosolenia gardineri* DENDY
5. *Leucosolenia laxa* KIRK
6. *Leucosolenia mutsu* HOZAWA

Family Sycettidae

7. *Sycetta quadriradiata* HOZAWA
8. *Sycon ornatum* KIRK
9. *Sycon misakiensis* HOZAWA

Family Heteropiidae

10. *Grantessa intusarticulata* (CARTER)
11. *Grantessa ampullae*, n. sp.

Family Grantiidae

12. *Paragrantia waguensis*, n. gen. and n. sp.
13. *Leucandra rigida*, n. sp.
14. *Leucandra spinosa*, n. sp.

15. *Leucandra glabra*, n. sp.
16. *Leucandra fragilis*, n. sp.
17. *Leucandra abratisbo* HOZAWA
18. *Leucandra mediocanellata*, n. sp.

As is seen from the above list, the number of species is 18 in all and they are referable to 6 genera belonging to 4 families.

Of the said 18 species, 10 are those previously known while the remaining 8 are described here for the first time.

Paragrantia is a genus newly erected to receive the new species of *Paragrantia waguensis*. *Paragrantia waguensis* is peculiar in having a skeletal structure composed of proper spicules around the apophyle of each flagellated chamber. Such features appearing in the apophyle skeleton do not seem to be present in any other calcareous sponges.

DESCRIPTION OF THE SPECIES

1. *Leucosolenia clathrata* (CARTER)

Leucetia clathrata CARTER, 1883, p. 33, Pl. I, figs. 13-17.

Clathrina tripodifera var. *gravida* CARTER, 1885-1886, p. 507.

Leucosolenia tripodifera var. *gravida* DENDY, 1891, p. 68.

Leucosolenia intermedia KIRK, 1895, p. 208, Pl. IV, fig. 2; BRØNSTED, 1926, p. 298.

Leucosolenia clathrata DENDY and ROW, 1913, p. 728; ROW and HOZAWA, 1931, p. 730.

There are many specimens of this species in the collection. They were all gathered on the shore at Wagu, Miye Prefecture. Each of these specimens consists of colonies of flattened and anastomosing ascon-people attached to the algae firmly.

This species was first described by CARTER (1883) in the name of *Leucetia clathrata* and afterwards it was recorded by several investigators. As ROW and HOZAWA (1931) have fully reported on this species, there is no need to add any further notes.

Previously known Distribution.— S. W. coast of Australia (CARTER); Near Port Phillip Heads, Westernport (Victoria), Kent Islands (Bass Strait) (DENDY); Cook Strait (KIRK); Island Bay, Wellington, N. Z. (BRØNSTED); Geraldton District, Fremantle Bay, Bunbury Bay (ROW and HOZAWA).

Locality and Register Nos. of Specimens.— Wagu, Miye Prefecture, A 1, A 2, A 3.

2. *Leucosolonia ventosa*, n. sp.

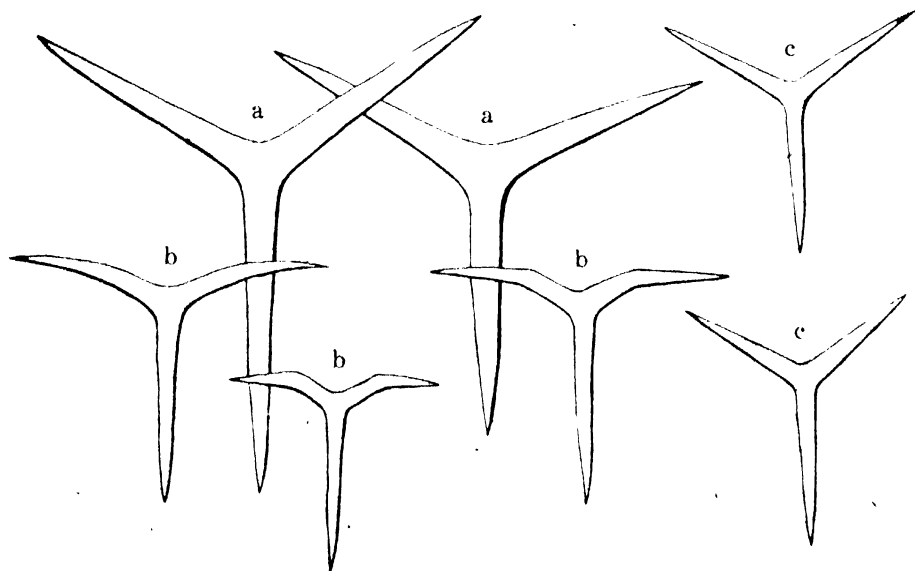
(Pl. IV, fig. 1; textfig. 1)

The unique specimen (Spec. No. A 1) representing this new species was obtained at Wagu. The sponge is irregularly massive in shape and is more or less laterally compressed, being attached to some seaweed. The sponge is rather small and attains to a length of about 8 mm. The surface of the sponge is perforated by numerous minute pseudopores. The pseudopores are circular in outline with a diameter of $160\ \mu$.

The colour of the specimen in alcohol is white and the texture is slightly rigid.

Structures. The skeleton is made up of triradiates only. There are, however, two different types of triradiates, the larger and the smaller. The larger triradiates are confined to the outermost parts of the sponge-colony, being rather closely set all over the pseudodermal surface, but leaving circular pseudopores. The smaller ones, which form the main skeleton supporting the remaining part of the sponge body, are arranged irregularly in several layers.

The wall of the ascon-tubes contains only the smaller triradiates which are arranged in a thin layer.



Textfig. 1. *Leucosolonia ventosa*, n. sp. a, Large triradiates. b, Small triradiates. c, Triradiates of ascon-tube. (All $\times 200$)

Spicules (Textfig. 1).—Large triradiates of pseudoderm (a) slightly sagittal. All rays are straight and are of nearly equal thickness being $20\text{--}25\mu$. The basal ray is slightly longer than paired rays, being $150\text{--}180\mu$ long. Paired rays equal and $140\text{--}150\mu$ long.

Small triradiates of the same (b) also slightly sagittal, all rays being nearly equally thick. The basal ray straight, gradually tapering to a sharp point, longer than paired rays, $100\text{--}120\mu$ long and $10\text{--}14\mu$ thick at base. Paired rays equal, straight or slightly curved forwards, or distinctly curved at a point shortly distant from base, $70\text{--}90\mu$ long and $10\text{--}14\mu$ thick at base.

Triradiates of ascon-tubes (c) slightly sagittal, much smaller than those of pseudoderm. All rays straight, gradually and sharply pointed, and are of equal thickness of 10μ at base. The basal ray slightly longer than paired rays, $100\text{--}120\mu$ long. Paired rays equal and $85\text{--}100\mu$ long.

Locality.—Wagu, Miye Prefecture.

Remarks.—This species appears to closely resemble MICHLUCHO-MACLAY's *Leucosolenia blanca*¹⁾ and HAECKEL's *L. phillipina*,²⁾ but differs from them, however, in the proportion of the basal ray to paired rays of large and small triradiates, that is, the basal rays of *L. blanca* and *L. phillipina* are much longer than the paired rays. And, moreover, the paired rays of these species are always straight. While in the species under observation the basal ray is slightly longer than the paired rays which are straight or curved.

This species may be easily distinguished from von LENDENFELD's *L. macleayi*³⁾ and DENDY's *L. stipitata*⁴⁾ by the absence of peduncle and the difference in arrangement of spicules.

3. *Leucosolenia reticulum* (O. SCHMIDT)

(Textfig. 2)

Nardoa reticulum O. SCHMIDT, 1862, p. 18, Taf. I., figs. 8-8 c.

Nardopsis reticulum HAECKEL, 1870, p. 247.

Tarrus reticulatus HAECKEL, 1870, p. 244.

Ascandra reticulum HAECKEL, 1872, Bd. II., p. 87; Bd. III., Taf. XIV, XX; von LENDENFELD, 1891, p. 223, Taf. VIII., figs. 7, 15.

¹⁾ *Guancha blanca*, MICHLUCHO-MACLAY, 1868,

²⁾ *Ascetta blanca* var. *phillipina*, HAECKEL, 1872, Bd. II. pp. 38-40; Bd. III, Taf. V., figs. 5 a-5 c.

³⁾ *Ascetta macleayi*, von LENDENFELD, 1885, pp. 1086-1087, Pl. 62, figs. 11-13.

⁴⁾ *Leucosolenia stipitata*, DENDY, 1891, pp. 51-52, Pl. I., figs. 4-6; Pl. IV., fig. 2; Pl. IX., fig. 5.

Clathrina reticulum MINCHIN, 1896,

Leucosolenia reticulum DENDY and ROW, 1913, p. 723; BREITFUSS, 1932, p. 243; 1935, p. 14; TOPSENT, 1936, p. 22, figs. 10, 11.

Of this species there is a single specimen (Spec. No. A 5) in the collection. It was secured by the present writer at Wagu, Miye Prefecture.

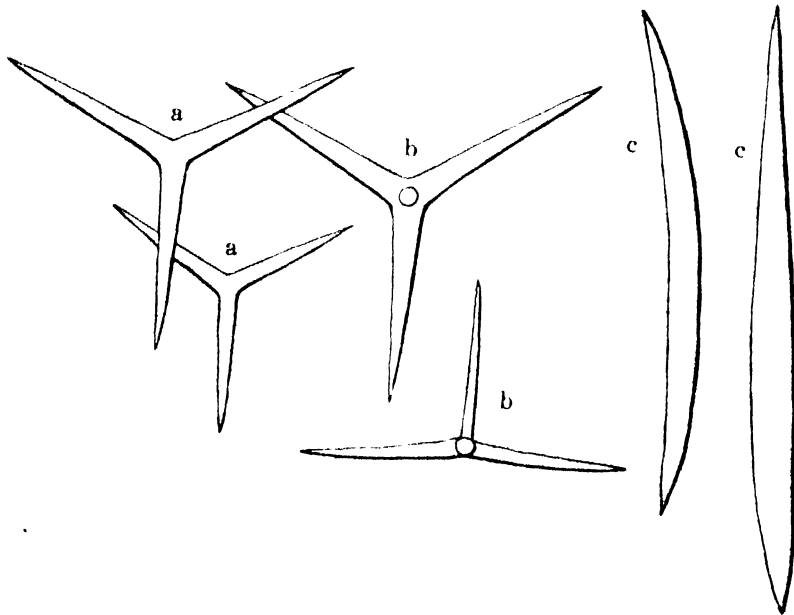
The sponge forms an irregularly shaped mass attached directly to the substratum. The height of the entire specimen is about 10 mm. and the greatest breadth is 4 mm. The pseudopores are evenly distributed over the pseudoderm.

The colour is white in alcohol and the texture is rigid.

Structure.—The skeleton is composed of triradiates, quadri-radiates, and oxea. The triradiates are arranged in a confused layer, the spicules being placed close together with rays overlapping. The oxea occur here and there and project more or less outwards through the pseudodermal surface.

The wall of ascon-tubes consists mainly of quadri-radiates, whose slender apical rays as usual projecting into the gastral cavity.

Spicules (Textfig. 2).—Triradiates (a) regular, with straight and sharply pointed rays, 80–130 μ long and 10–14 μ thick at base.



Textfig. 2. *Leucosolenia reticulum* (O. SCHMIDT). a, Triradiates. b, Quadri-radiates. c, Oxea. (All $\times 200$)

Quadriradiates (b) are of nearly the same size and shape as the tri-radiates above mentioned, but with the addition of an apical ray. The apical ray is straight and slender, gradually and sharply pointed, projecting freely into the gastral cavity. It is slightly thinner and longer than the facial rays, 90-150 μ long and 6-8 μ thick at base.

Oxea (c) nearly straight or slightly curved, sharply pointed at both ends, but one end is usually slender and more finely pointed than the other. Oxea is even in outline, 230-360 μ long and 18-26 μ thick at the broadest portion.

Remarks.—The specimen from Wagu seem to agree well in features with a species which was first described by O. SCHMIDT (1862) and afterwards has been very fully recorded by HAECKEL (1872) and VON LENDENFELD (1891). The dimensions of spicules in the specimen to hand seem to be a little larger than those mentioned by HAECKEL and VON LENDENFELD and, moreover, no S-shaped oxea can be found in the writer's specimen. These differences between them may be looked upon as variations. Thus the writer is inclined to identify the specimen with HAECKEL, VON LENDENFELD's species.

In 1935, BREITFUSS reported on this species from the Adria Sea. In that report he stated that the *Leucosolenia sagamiana*¹⁾ described by the present writer to be synonymous with his own species without giving his grounds for this opinion. But his conclusion seems unconvincing, because *L. sagamiana* differs from BREITFUSS' species in several particulars, viz. in *L. sagamiana* 1) the quadriradiates are more numerous than the tri-radiates, 2) the apical ray is much longer than the facial rays, that is the former is about twice as long as the latter. While in *L. reticulum*, the apical ray is nearly equal in length to the facial rays. 3) Oxea of *L. sagamiana* is very large in comparison with the triradiates. The tri-radiates are nearly equal in size in both *L. sagamiana* and *L. reticulum*. These differences above alluded to occur rather constantly, and thus it seems to be reasonable to distinguish these *L. sagamiana* and *reticulum* as two quite separate species.

Previously known Distribution.—Adria Sea (O. SCHMIDT, HAECKEL, VON LENDENFELD, BREITFUSS); Greenland (O. SCHMIDT); New Foundland (BREITFUSS); Banyuls (TOPSENT); Napoli (VOSMAER).

Locality and Register No. of Specimen.—Wagu, Miye Precture, A 5.

¹⁾ *Leucosolenia sagamiana*, HÓZAWA, 1929, pp. 281-282, Pl. I., figs. 1, 2; Textfig. 1.

4. *Leucosolenia gardineri* DENDY

Leucosolenia gardineri DENDY, 1913, p. 2, Pl. I., figs. 1-2, Pl. III., figs. 1-3.

This species is represented in the collection by seven specimens, which were obtained at Wajima, Noto Province.

The sponge body shows a reticulation very closely-meshed, and more or less flattened, attached to the substratum directly. The surface of the sponge is perforated by numerous pseudopores with a diameter of 200-400 μ .

In respect to the canal system and skeletal arrangement, these specimens are exactly identical with the type specimen. In spiculation, these specimens are also identical with the type, except as regards the thickness of the spicules. Namely, the thickness of the spicules in the specimens to hand is 10-22 μ , while that of the type specimen is 7-12 μ . But this difference may be considered as a variation in spicules.

Previously known Distribution. Chagos Archipelago (DENDY).

Locality and Register No. of Specimens.—Wajima, Province Noto, B 1.

5. *Leucosolenia laxa* KIRK

Leucosolenia laxa KIRK, 1895, p. 208, Pl. IV., fig. 1; DENDY and ROW, 1913, p. 722; HOZAWA, 1928, p. 220, Pl. I., figs. 4, 5.

This species is represented in the collection by many specimens obtained from two different localities. The first group of specimen (Spec. No. B 2) was collected from Wajima by the present writer. The other group was obtained at Oshima, Miyagi Prefecture. All these specimens are irregular and massive in shape being attached to the substratum directly. The specimens vary in size from a very minute example up to a mass 15 mm. long and 20 mm. wide.

In anatomical structure these specimens agree well with the description given by KIRK.

Previously known Distribution.—New Zealand (KIRK); Mutsu Bay (HÔZAWA).

Localities and Register Nos. of Specimens.—Wajima, Ishikawa Prefecture, B 2; Oshima, Miyagi Prefecture, C 1.

6. *Leucosolenia mutsu* HOZAWA

Leucosolenia mutsu HOZAWA, 1928, p. 219, Pl. I., figs. 1-3.

There are two specimens of this species in the collection. They were taken at Oshima in Miyagi Prefecture. Each of these specimens forms an irregular mass consisting of a loose net-work of ascon-tubes. One specimen attains to the length of 7 mm. and the other to 5 mm.

In the general structure of the ascon-tubes, in the arrangement of the skeleton and in the spiculation, these specimens are identical with those of the type.

Previously known Distribution.—Mutsu Bay (HOZAWA).

Locality and Register No. of Specimens. Oshima, Miyagi Prefecture, C 2.

7. *Sycetta quadriradiata* HOZAWA

Sycetta quadriradiata HOZAWA, 1929, p. 294, Pl. II., figs. 12, 13, Textfig. 7.

This species is represented by a single specimen (Spec. No. A 6) in the collection. It was obtained from Wagu, Miye Prefecture. The specimen is a solitary individual of tubular form being about 10 mm. in total length and 1.5 mm. in the greatest breadth. The colour in alcohol is greyish white and the texture is soft.

With regard to the arrangement and the spiculation, the specimen is exactly similar to the type specimen.

Previously known Distribution. Off Yamakawa, Kagoshima Bay (HÔZAWA).

Locality and Register No. of Specimen. Wagu, Miye Prefecture, A 6.

8. *Sycon ornatum* KIRK

Sycon ornatum KIRK, 1897, p. 314, Pl. 31, figs. II a, II b; Pl. 32, fig. II; DENDY and ROW, 1913, p. 747.

This species is represented by five specimens (Spec. No. C 3), all of which being obtained at Oshima, Miyagi Prefecture.

They are all of a nearly similar appearance. Each of them represents a solitary individual of an elongated cylindrical form, broadest at a part a little below the top, and narrowing towards the base. The sponge is attached to the substratum by means of its base and shows at the upper end an osculum, which is surrounded by a well-developed collar. In the largest specimen the total length is 11 mm. and the greatest breadth is 2.5 mm.

The species has been fully described by KIRK (1897), so that no further details are necessary.

Previously known Distribution.—Cook Strait (KIRK).

Locality and Register No. of Specimens.—Oshima, Miyagi Prefecture, C 3.

9. *Sycon misakiensis* HOZAWA

Sycon misakiensis HOZAWA, 1929, p. 300, Pl. III., figs 16, 17, Textfig. 9.

This species is represented by a single specimen (Spec. No. C 4) in the collection. It was obtained at Oshima, Miyagi Prefecture.

The sponge represents a solitary individual of a slightly laterally compressed elongated sac-shape, attached by the narrowed base to the substratum. The breadth of the specimen is about 5 mm. and the thickness of the wall is 1.5 mm. in the middle part. The upper part of the sponge was broken, so that it is impossible either to ascertain the features of oscular margin or to measure the height. But, as in the external appearance and in the internal structure it agrees well with the type specimen, the writer identified it with the present species.

Previously known Distribution.—Misaki (HÓZAWA).

Locality and Register No. of Specimen.—Oshima, Miyagi Prefecture, C 4.

10. *Grantessa intusarticulata* (CARTER)

Hypograntia intusarticulata CARTER, 1885-1886, p. 45.

Hypograntia medioarticulata CARTER, 1885-1886, p. 46.

Grantessa intusarticulata DENDY, 1892, p. 108; 1893, p. 181, p. 201, Pl. XIII., fig. 18; DENDY and ROW, 1913, p. 753; HOZAWA, 1916, p. 14, Pl. I., fig. 4, Pl. II., fig. 13, Textfig. 3; 1929, p. 318; 1932, p. 7; BRØNSTED, 1926, p. 308; ROW and HOZAWA, 1931, p. 775.

Grantia intusarticulata BREITFUSS, 1897, p. 219.

Many specimens of this species were collected at Wagu and at Wajima. All the specimens represent a colony of several small and tubular individuals joined together at their bases.

The colonies from Wagu consist of thicker tubes than in those from Wajima, and moreover the oscula are surrounded by a fringe of well-developed oxea. The colour in alcohol is white. But in the specimens from Wajima, the oscular fringes are not developed so well.

In features, both of the specimens from Wagu and from Wajima agree well with the description of this species given by previous writers.

Previously known Distribution.—Near Port Phillip Heads (CARTER, DENDY); Watson's Bay, Port Jackson (DENDY); Sagami Sea, Japan

(HÔZAWA); Island Bay, Wellington, N. Z. (BRØNDSTED); Geraldton District, S. W. Australia (ROW and HÔZAWA).

Localities and Register Nos. of Specimens.—Wagu, Miye Prefecture, A 7, A 8, A 9, A 10; Wajima, Ishikawa Prefecture, B 3.

11. *Grantessa ampullae*, n. sp.

(Pl. IV, fig. 2; Textfig. 3)

This species is based upon a single specimen (Spec. No. B 4) which was secured at Wajima, Noto. It represents a cup-like individual laterally compressed. It measures about 5.5 mm. long by 7 mm. broad at the widest part and the thickness of the wall is about 0.8 mm. The osculum is circular in shape with a diameter of 2 mm. and its margin is thin and naked without any fringe. The dermal and the gastral surfaces are both smooth without any projecting spicules. The colour of the sponge in alcohol is greyish white and the texture is rigid.

Structure.—The canal system is typically syconoid. The flagellate chambers are arranged radially with regularity. They are straight, circular in transverse section and are usually undivided.

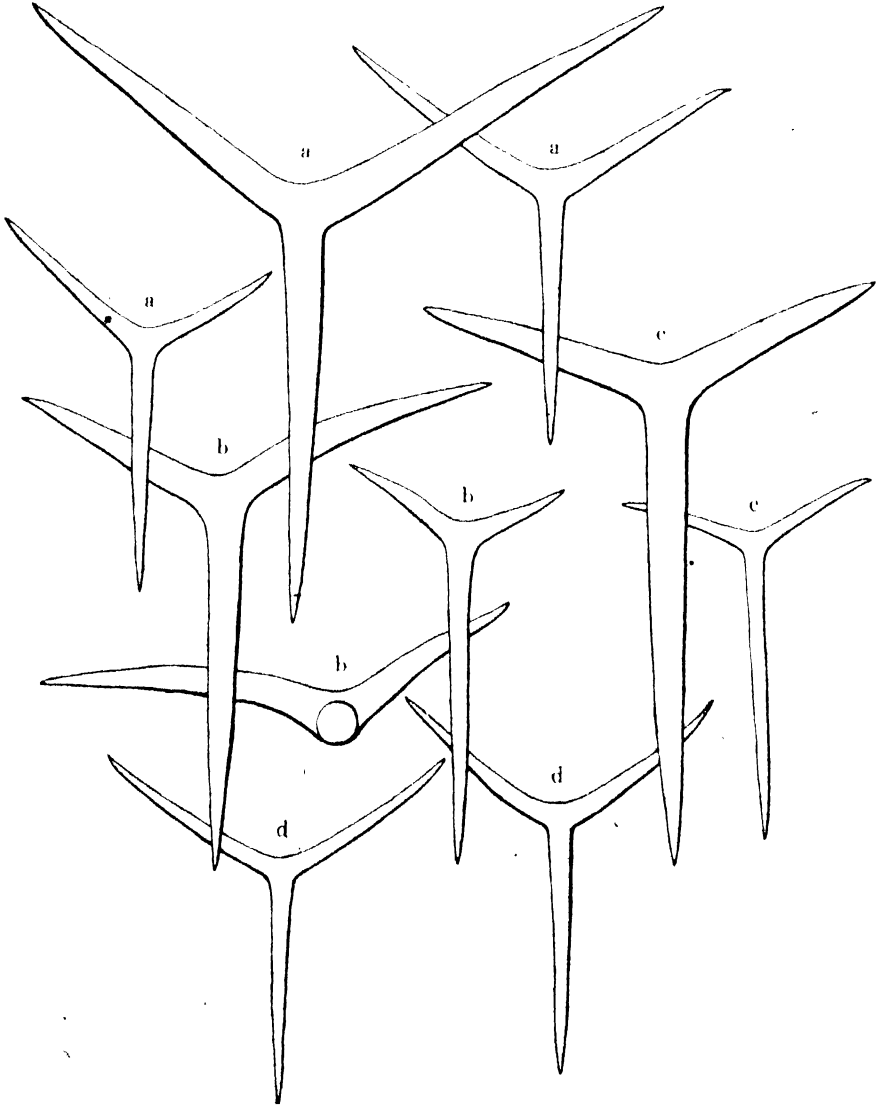
The dermal skeleton is composed of densely and tangentially placed sagittal triradiates and of the paired rays of subdermal pseudosagittal triradiates. In general there is not any regularity in the orientation of the dermal triradiates. The tubar skeleton is made up of the strong centripetal basal rays of subdermal pseudosagittal triradiates and of the centrifugal basal rays of subgastral triradiates. The gastral skeleton is more weakly developed than the dermal and is composed of tangentially placed slender gastral triradiates and of strongly developed paired rays of subgastral triradiates. The osculum has no special skeleton.

Spicules (Textfig. 3).—Dermal triradiates (a) sagittal. All rays are of equal thickness being $14\text{--}32\ \mu$, tapering from base to a sharp point. Basal ray straight, $135\text{--}320\ \mu$ long. Paired rays are in most cases unequal and are either almost straight or slightly curved. The longer ray of the same is nearly equal to the basal ray in length, $135\text{--}330\ \mu$ long. The shorter ray is almost straight or slightly curved near the sharp point, $90\text{--}270\ \mu$ long.

Subdermal triradiates (b) pseudosagittal, stout. All rays are of different length and lying not in one plane, but are of nearly the same thickness. Basal ray much longer than paired rays, sharp-pointed, straight or slightly curved, $260\text{--}480\ \mu$ long and $20\text{--}40\ \mu$ thick at base. Paired rays are of

different length and shape. The longer ray of the same is strongly curved backward and gradually sharp-pointed, 100–250 μ long and 20–40 μ thick at base. The shorter ray is almost straight and 90–240 μ long.

Subgastral triradiates (c) strongly sagittal, stout. All rays are equally thick and smooth. Basal ray straight, gradually tapering from base to



Textfig. 3. *Grantessa ampullae*, n. sp. a, Dermal triradiates. b, Subdermal pseudo-sagittal triradiates. c, Subgastral triradiates. d, Gastral triradiates. (All $\times 150$).

sharp point, considerably longer than the paired rays, 260–410 μ long and 14–30 μ thick at base. Paired rays nearly equal in length, straight or slightly curved and lying not in one plane with basal ray, 90–250 μ long and 14–30 μ thick at base.

Gastral triradiates (d) sagittal and slender. Basal ray straight, sharply pointed, 90–210 μ long and 10–18 μ thick at base. Paired rays nearly equal, always curved forwards and not lying in the same plane with basal ray, 65–190 μ long and 10–18 μ thick at base.

Remarks.— In the structure of the skeleton, this species bears a close resemblance to JENKIN's *Grantessa simplex*¹⁾ and to the present writer's *G. basipapillata*,²⁾ but from the former it differs in the unequal length of paired rays of the dermal triradiates, and from the latter in the absence of the gastral quadriradiates. Moreover, this species differs from both of these species in the external appearance.

Locality and Register No. of Specimen.— Wajima, Ishikawa Prefecture, B 4.

12. *Paragrantia waguensis*, n. gen. and n. sp.

(Pl. V, figs. 8–11; Textfig. 4)

There are eight specimens of this new species in the collection, which are now deposited in the museum of the Tōhoku Imperial University. All of these specimens were obtained by the writer at Wagu, Miye Prefecture, in July, 1933.

The sponge (Pl. V, fig. 8) forms a rather simply branched colony. It grows vertically upwards, being attached directly to the substratum by means of its base. The largest specimen is about 40 mm. long and is 8 mm. in the greatest breadth. The dermal surface of the body is nearly smooth without any projecting oxea, but the gastral is punctated on account of numerous small exhalant pores. The sponge wall is about 2 mm. thick in the middle part of the body, gradually becoming thinner towards the osculum. The osculum is elliptical and is surrounded by a feebly developed collar. The osculum is 3.5 mm. long and 2 mm. wide.

The texture is fairly rigid. The colour of the sponge in alcohol is milky white.

Structure.— The canal system is of syconoid. The flagellate chambers are of an elongated sac-like shape, measuring about 1.5 mm. in length

¹⁾ *Grantessa simplex*, JENKIN, 1908, pp. 446–449; Textfigs. 93–97.

²⁾ *Grantessa basipapillata*, HÔZAWA, 1916, pp. 19–23, Pl. I., fig. 6; Pl. II., fig. 14; Textfig. 4.

and about $150\ \mu$ in diameter. They are arranged radially around the gastral cavity and usually are not branched.

The skeleton (Pl. V, fig. 9) is composed of triradiates, quadriradiates and oxea, the last mentioned spicules occurring only in the oscular margin. The dermal skeleton is consisted of triradiates, which are tangentially arranged in dense several layers.

The tubar skeleton is of an articulate type and is made up of triradiates set in many rather confused layers. It receives an addition of some basal rays of subgastral triradiates.

The gastral skeleton is composed of tangentially placed tri- and quadriradiates, which are provided with apical rays projecting into the gastral cavity.

Around each apopyle of the flagellate chamber, there may be distinguished a distinct skeletal structure. This apopyle skeleton is composed of a number of quadriradiates, which are arranged in several layers and which are set radially around each apopyle with their apical rays projecting into the apopyle (Pl. V, fig. 10).

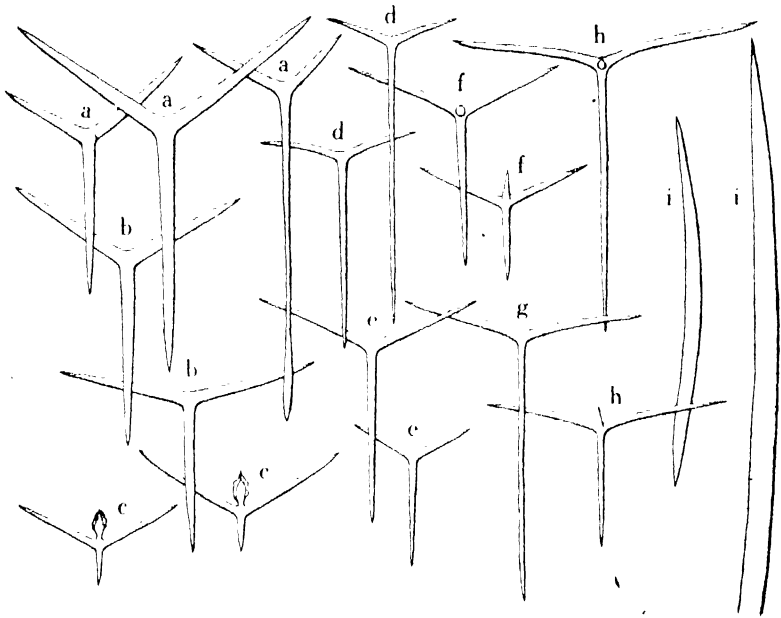
The skeleton of the oscular margin is made up of tri- and quadriradiates with an addition of some oxea. The oxea run longitudinally and parallel with one other, their distal ends almost not projecting from the oscular margin.

Spicules (Textfig. 4).— Dermal triradiates (a) sagittal and are of two kinds. In one kind of the spicules, all rays are nearly equally thick, measuring $12\text{--}20\ \mu$ at base, gradually tapering to sharp point, and lying in one plane. Basal ray straight, always longer than paired rays, $80\text{--}320\ \mu$ long. Paired rays slightly unequal, either straight or slightly curved forwards. The longer ray is $130\text{--}220\ \mu$ long and the shorter is $60\text{--}170\ \mu$ long. The other kind of the spicules are very few in number in comparison with the former. Basal ray nearly straight, but is narrowed in the middle part becoming thinner than the end. They are $250\text{--}420\ \mu$ long and the thickness measured at base, in the middle, and at end are respectively $10\text{--}16\ \mu$, $4\text{--}5\ \mu$, and $8\text{--}10\ \mu$. Paired rays unequal and the difference in length is rather distinct. The shorter ray straight, $50\text{--}110\ \mu$ long and $10\text{--}16\ \mu$ thick at base. The longer ray also straight, tapering from base to the sharp point, $80\text{--}135\ \mu$ long and $10\text{--}16\ \mu$ thick at base.

Tubar triradiates (b) sagittal. Basal ray straight, longer than paired rays, $140\text{--}225\ \mu$ long and $10\text{--}16\ \mu$ thick at base. Paired rays equal, $90\text{--}160\ \mu$ long and $10\text{--}16\ \mu$ thick at base.

Quadriradiates of apopyle (Pl. V, fig. 11; Textfig. 4 c) have the both

of the paired and basal rays gradually tapering and sharply pointed and of nearly equal thickness. All rays not lying completely in one plane. Basal ray straight and short, 20–25 μ long and 4 μ thick at base. Paired rays equal, more or less curved around the apophyle, 56–70 μ long and 4 μ thick at base. The apical ray is broadest in the middle part which is surrounded by an irregularly undulating outline, and whence it is narrowed towards the base and the pointed end. As a whole the apical ray of the apophyle quadriradiate spicule is peculiar in appearance looking like a frame of torch. It is about 20 μ long and 12–18 μ thick at the broadest part.



Textfig. 4. *Paragraptia waguensis*, n. gen. and sp. a, Dermal triradiates. b, Tubar triradiates. c, Quadriradiates of apophyle. d, Subgastral triradiates. e, Gasteral triradiates. f, Gasteral quadriradiates. g, Triradiates of oscular margin. h, Quadriradiates of oscular margin. i, Oxea. (a, b, d–i $\times 100$, c $\times 200$)

Subgastral triradiates (d) sagittal. All rays slender, equally thick, tapering to sharp point. Basal ray straight, much longer than paired rays, 180–350 μ long and 8–10 μ thick at base. Paired rays equal, widely divergent, nearly forming a straight line, otherwise they are slightly curved at base, 65–110 μ long.

Gasteral triradiates (e) strongly sagittal. Basal ray straight, slender,

100–390 μ long and 8–10 μ thick at base. Paired rays 70–200 μ long and 8–10 μ thick at base.

Gastral quadriradiates (f) nearly similar to the above gastral triradiates, only differing in the presence of an apical ray. Basal ray 100–390 μ long and about 10 μ thick at base. Paired rays 115–170 μ long and about 10 μ thick at base. Apical ray is 40–90 μ long and about 12 μ thick at base.

Triradiates of oscular margin (g) strongly sagittal, with basal ray straight, much longer and thinner than paired rays, about 340 μ long and 10 μ thick at base. Paired rays equal, nearly straight, about 150 μ long and 12 μ thick at base.

Quadriradiates of oscular margin (h) strongly sagittal. As in the case of the oscular triradiates, the basal ray is straight and is longer and thinner than paired rays, 150–420 μ long and about 8 μ thick at base. Paired rays equal, slightly doubly curved, first backwards and then slightly forwards, 150–200 μ long and 10 μ thick at base. Apical ray straight and short, 20–40 μ long and 10 μ thick at base.

Oxea of oscular margin (i) elongate spindle shaped, slightly curved, sharply pointed at both ends, even in outline, 300–880 μ long and 14–28 μ thick in the thickest portion.

Locality.—Wagu, Miye Prefecture.

Remarks.—Judging from the features above mentioned, this sponge without doubt belongs to the family Grantiidae. But the most conspicuous feature of this sponge is the presence of a distinct apopyle skeleton, which is formed by quadriradiates of peculiar features around the apopyle. So far as the present writer is aware, such a structure, composed of spicules of this peculiar nature, has not hitherto been observed in the Grantiidae. It seems to him to be highly reasonable to create a new genus *Paragrastia* among the family Grantiidae to include this new species, basing the generic diagnosis upon the most peculiar features of the apopyle skeleton above alluded to. The genus *Paragrastia* is diagnosed as follows:

Paragrastia, n. gen.

“Canal system syconoid. Sponge usually a simple branched colony. Tubar skeleton articulate, composed of radiate spicules, which may or may not be supplemented by oxea. Around each apopyle of the flagellated chamber there exists a special skeleton composed of proper radiates.”

13. *Leucandra rigida*, n. sp.

(Pl. IV, fig. 3; Textfig. 5)

This new species is represented in the collection by three specimens. Each of them forms a massive colony with a number of oscula. The writer has selected the largest specimen shown in Pl. IV, fig. 3 on which to base the following detailed description.

The sponge is in the form of a mass with three oscula. The mass is broadest near the base and is narrowed towards one of the oscula. The total length of the body is about 20 mm. and the greatest breadth is about 15 mm. The oscula are circular or oval in shape with a diameter of 1.5–2 mm. and are not surrounded by any fringe. The dermal surface is hispid due to the projecting oxea, and the gastral surface is also slightly hispid from the apical rays of the gastral quadriradiates. The colour in alcohol is white and the texture is rigid.

Structures.—The canal system is of the leuconoid type. The flagellate chambers are of sac-like shape, circular or oval in cross-section, measuring up to $140\ \mu$ long by $90\ \mu$ thick. The exhalant canals are $300\text{--}640\ \mu$ in diameter.

The dermal skeleton is composed of triradiates, large oxea and microxea. The triradiates are placed tangentially and are closely set in several layers without any orientation. The large oxea occur in the sponge wall with their distal ends projecting from the dermal surface to some extent. The microxea are in sparse distribution, each projecting nearly vertically from the dermal surface.

The tubar skeleton is made up of triradiates which vary in size and are irregularly scattered in the chamber layer.

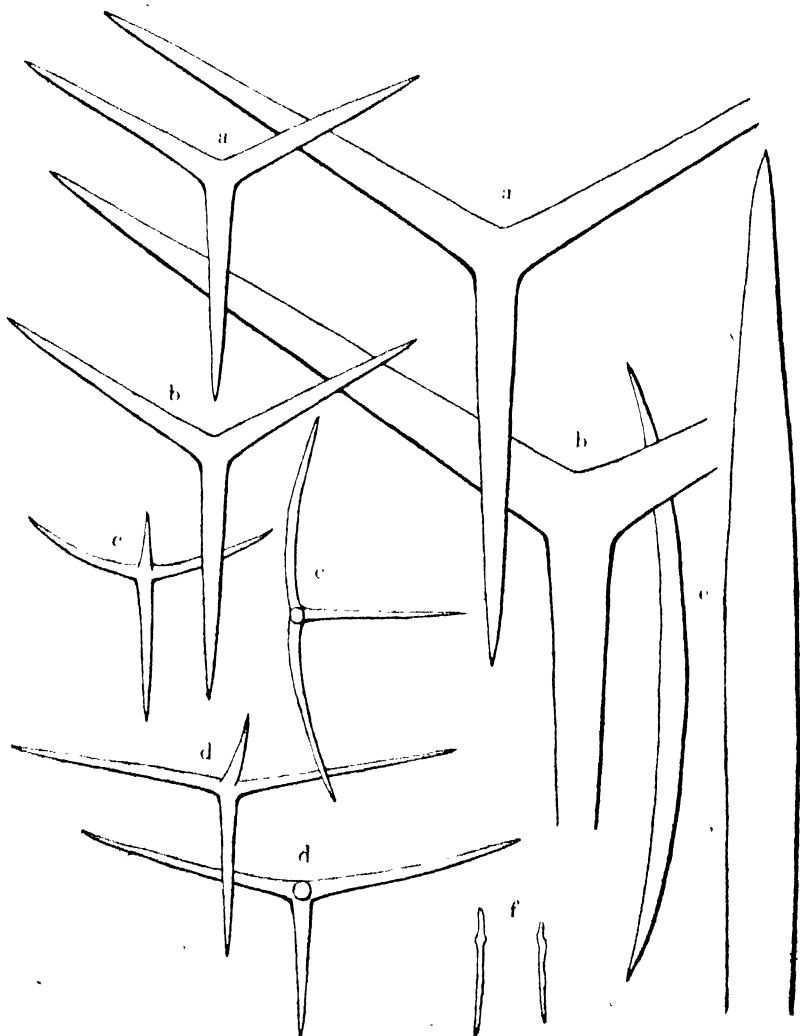
The gastral skeleton consists of microxea and of slender quadriradiates. The quadriradiates are fairly closely set and are tangentially disposed in a few layers with their apical rays projecting into the gastral cavity as well as into the exhalant apertures.

The oscular skeleton is composed of dermal and gastral spicules, and there is not any peculiarity to be mentioned of their arrangement.

Spicules (Textfig. 5).—Dermal triradiates (a) regular or subregular, variable in size. Rays equally thick, even in outline, sharply pointed, $100\text{--}770\ \mu$ long and $10\text{--}60\ \mu$ thick at base.

Tubar triradiates (b) regular, varying in size, stouter than the triradiates of the dermal skeleton. Rays straight, tapering from the base to sharp point, equally thick, $200\text{--}900\ \mu$ long and $20\text{--}90\ \mu$ thick at base.

Quadriradiates of the larger exhalant canals (c) sagittal and slender. Basal ray straight nearly as thick as the paired rays but shorter. It is $100\text{--}140\ \mu$ long and $8\text{--}10\ \mu$ thick at base. Paired rays equal, slightly curved forwards, $140\text{--}210\ \mu$ long and $8\text{--}10\ \mu$ thick at base. Apical ray straight, variable in length, slightly thinner than facial rays, $60\text{--}310\ \mu$ long and $6\text{--}8\ \mu$ thick at base.



Textfig. 5. *Leucandra rigida*, n. sp. a, Dermal triradiates. b, Tubar triradiates. c, Quadriradiates of larger exhalant canal. d, Gastral quadriradiates. e, Large oxea. f, Gastral microxea. (a-e $\times 80$. f $\times 320$)

Gastral quadriradiates (d) sagittal. Rays rather slender and equally thick. Basal ray straight, shorter than paired rays, 70–140 μ long and 8–12 μ thick at base. Paired rays equal, slightly curved forwards, 100–270 μ long. Apical ray curved upwards, nearly as long as basal ray, 80–140 μ long.

Large oxea (e) spindle shaped, more or less curved, sharply pointed at both ends, 260–900 μ long and 10–38 μ thick at the thickest portion.

Dermal microxea (f) nearly straight. Close to one end occurs a nodiform ring. They measure about 40 μ in length and 2 μ in thickness.

Gastral microxea are similar to the dermal microxea in shape, 60–70 μ long and 3–4 μ thick.

Remarks.—This species seems to be closely related to the writer's *Leucandra solida*¹ obtained from the Sagami Sea, but it may be easily distinguished from the latter by the difference in spiculation. In the present species the large oxea have no nodiform ring and the dermal microxea are not spined. Moreover, the difference in size between the tubar and dermal triradiates is marked in the present species.

Locality and Register No. of Specimens.—Wagu, Miye Prefecture, A 13.

14. *Leucandra spinosa*, n. sp.

(Pl. IV, fig. 4; Textfig. 6)

The collection contains many specimens of this new species (Spec. No. A 14, A 15) which were obtained at Wagu, Miye Prefecture.

The sponges are more or less different from one another in size and shape, but are practically identical in the internal structure. The sponge represents a solitary individual of an oval, spherical or irregular shape, showing on the upper surface a single osculum which is surrounded by a well-developed collar. The dermal surface is hispid due to the projecting oxea. The gastral surface appears smooth but is perforated by numerous circular or oval apertures of exhalant canals. The colour in alcohol is white, the texture is compact and rather hard.

The largest specimen which the writer selected as the type of the species (Pl. IV, fig. 4) is oval in shape with a height of about 18 mm. and a breadth of 15 mm. It has a circular osculum provided with a well-developed collar, which is about 4 mm. in height and about 1.5 mm. in diameter. The gastral cavity is rather narrow and is irregular in shape. The sponge wall is about 8 mm. in the thickest parts.

¹⁾ *Leucandra solida*, HÔZAWA, 1929, pp. 362–365, Pl. X., figs. 59, 60; Textfig. 30.

The following description is based upon the type specimen.

Structures.—The canal system is typically leuconoid. The flagellate chambers are closely distributed in the chamber layer. They are oval shape or more or less elongate sac-shape, measuring up to 100μ long by 70μ thick.

The dermal skeleton is composed of slightly sagittal triradiates, which are tangentially arranged in several layers, and of two kinds of oxea. The large oxea occur in nearly vertical disposition to the dermal surface and their distal parts project far beyond the surface, while their proximal parts are embedded deep into the wall. The microxea occur fairly densely in vertical disposition to the dermal surface.

The skeleton of the chamber layer is made up of the proximal parts of large oxea and of triradiates which are thickly distributed and irregularly set together. The walls of the larger exhalant canals are lined with quadriradiates with the apical rays projecting into the canal.

The gastral skeleton consists of tangential tri- and quadriradiates and of vertical microxea. The apical rays of gastral quadriradiates project into the gastral cavity.

The skeleton of the oscular margin is supported by triradiates and long linear spicules. The latter form a very dense fringe around the osculum.

Spicules (Textfig. 6).—Dermal triradiates (a) slightly sagittal. Basal ray straight, tapering from the base to sharp-point, slightly longer than paired rays, $90-250\mu$ long and $8-22\mu$ thick at base. Paired rays equal, slightly curved backwards or doubly curved, backwards in basal parts and forwards in remaining parts, $65-200\mu$ long and $8-22\mu$ thick at base.

Triradiates of chamber layer (b) also slightly sagittal. Rays equally thick. Basal ray straight, sharply pointed, $150-270\mu$ long and $18-25\mu$ thick at base. Paired rays nearly equal, always doubly curved $150-250\mu$ long.

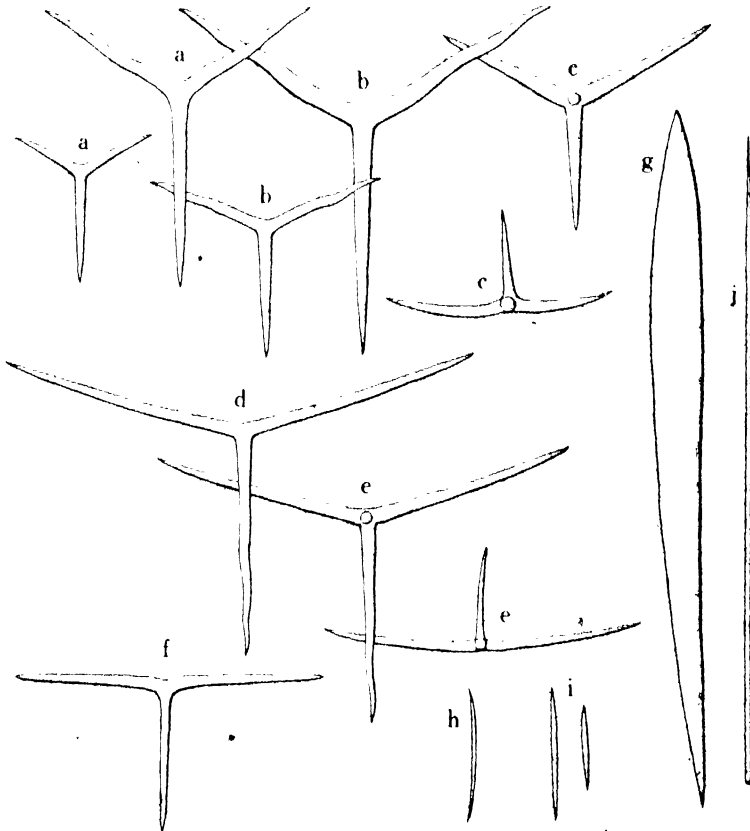
Quadriradiates of the larger exhalant canals (c) slightly sagittal. Basal ray straight, nearly as thick as paired rays but slightly shorter, $90-150\mu$ long and $16-20\mu$ thick at base. Paired rays equal, $130-170\mu$ long and $16-20\mu$ thick at base. Apical ray straight, sharply pointed, shorter and thinner than facial rays, $80-110\mu$ long and about 15μ thick at base.

Gastral triradiates (d) sagittal. Rays rather slender and equally thick. Basal ray slightly curved, shorter than paired rays, $160-270\mu$ long and $12-14\mu$ thick at base. Paired rays very widely divergent, slightly curved forwards, $190-330\mu$ long and $12-14\mu$ thick at base.

Gastral quadriradiates (e) almost similar to gastral triradiates, differing only in the presence of apical ray. Apical ray nearly straight, sharply pointed, shorter and thinner than facial rays, about $150\ \mu$ long and $8\text{--}10\ \mu$ thick at base.

Triradiates of oscular margin (f) strongly sagittal, being in the shape of figure T. Basal ray straight, sharply pointed, shorter and thinner than paired rays, $140\text{--}160\ \mu$ long and $8\text{--}12\ \mu$ thick at base. Paired rays equal, strongly divergent, $170\text{--}200\ \mu$ long and $15\text{--}20\ \mu$ thick at base.

Large oxea (g) spindle-shaped, broadest at a point nearer one end than the other, tapering towards both sharply pointed ends. They are



Textfig. 6. *Leucandra spinosa*, n. sp. a, Dermal triradiates. b, Tuber triradiates. c, Quadriradiates of larger exhalant canal. d, Gastral triradiates. e, Gastral quadriradiates. f, Triradiates of oscular margin. g, Large oxea. h, Dermal microoxea. i, Gastral microoxea. j, Linear spicule of oscular margin. (a-f $\times 100$, g, j $\times 50$, h, i $\times 200$)

straight and vary in length, $780\ \mu$ –1.8 mm. long and 50 – $110\ \mu$ thick in the thickest parts.

Microxea of dermal surface (h) slightly curved, sharply pointed at both ends, about $150\ \mu$ long and 2 – $4\ \mu$ thick in the middle parts.

Oxea of gastral surface (i) rather small, straight, spindle-shaped, and are sharply pointed at both ends. It measures 100 – $150\ \mu$ long and about $4\ \mu$ thick in the middle parts.

Linear spicules of oscular margin (j) very slender, straight, uniformly thick throughout their greater length and tapering towards the ends which are finely pointed, 2.8 – 4.5 mm. long and about $12\ \mu$ thick.

Remarks.—The above described species bears a marked resemblance to URBAN's *Leucandra apicalis*¹⁾ in external form and in canal system, but it differs from the latter in spiculation, especially in the form of triradiates and oxea.

Locality and Register Nos. of Specimens.—Wagu, Miye Prefecture; A 14, A 15.

15. *Leucandra glabra*, n. sp.

(Pl. IV, fig. 5; Textfig. 7)

This species is based upon a single specimen (Spec. No. A 16) which was secured at Wagu.

It consists of a massive piece of sponge approximately square shaped, measuring about 70 mm. by 50 mm. It is attached to the substratum by its base and there is a large hole in the central portion. Here and there a number of small oscula are seen scattered on the surface. Each osculum is naked and nearly circular in outline with a diameter of 1–2 mm. Both the dermal and gastral surfaces are smooth. The colour is white in alcohol and the texture is fairly rigid.

Structures.—The canal system is of leuconoid. The flagellate chambers which are closely packed in the chamber layer, are oval or spherical in form with a diameter of about $90\ \mu$. The exhalant canals are 240 – $620\ \mu$ in diameter.

The dermal skeleton is composed of regular triradiates only, and they are placed tangentially and are set closely in several layers.

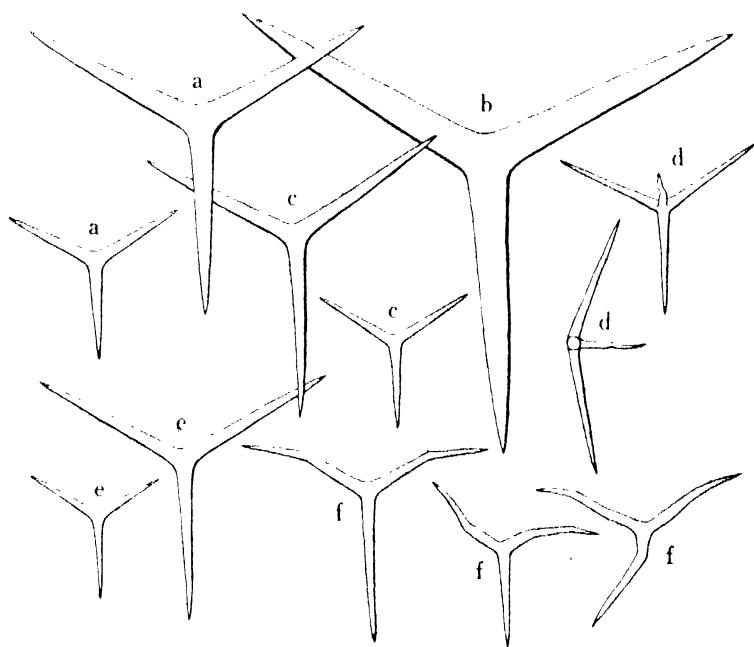
The skeleton of the chamber layer is made up of the following elements: 1) large regular triradiates which are scattered here and there in the chamber layer; 2) small triradiates, which are most numerous of

¹⁾ *Leucandra apicalis*, URBAN, 1905, pp. 67–70; Taf. VII., figs. 89–107.

all the elements found in the chamber layer; and 3) quadriradiates are few in number and are found scattered among the small triradiates.

The gastral skeleton is consisted of triradiates only. The skeleton of the oscular margin is composed of triradiates only, but the shape of the spicules varies from regular to irregular. The irregular ones seem to be modifications of regular spicules.

Spicules (Textfig. 7).—Dermal triradiates (a) regular. All rays equally thick tapering from base to the sharp point, 120–240 μ long and 14–28 μ thick at base.



Textfig. 7. *Leucandra glabra*, n. sp. a, Dermal triradiates. b, Large tubar triradiate. c, Small tubar triradiates. d, Tubar quadriradiates. e, Gastral triradiates. f, Irregular triradiates of oscular region. (All $\times 100$)

Large tubar triradiates (b) regular and very stout. Rays straight, sharply pointed, 400–950 μ long and 42–110 μ thick at base.

Small tubar triradiates (c) almost similar to the large tubar triradiates in shape, but are smaller and are slender, 100–200 μ long and 10–20 μ thick at base.

Tubar quadriradiates (d) almost like small tubar triradiates with addition of an apical ray. Apical ray curved abruptly, sharply pointed, shorter

and thinner than facial rays, about 80μ long and 10μ thick at base.

Gastral triradiates (e) similar to the small triradiates. It measures $90-200\mu$ in length and $12-20\mu$ in thickness.

Triradiates of the oscular margin (f) regular or irregular. Regular triradiates are nearly the same in shape and size with the gastral triradiates. Irregular ones are modified from the regular triradiates. Rays are nearly equally thick and equal in length. Most of these spicules have one ray straight and two curved. Of the curved rays the one curved forwards and the other backwards, or both are curved backwards. Sometimes all of the rays are curved and thus the spicules become very irregular.

Remarks.—This species seems to be quite distinct from the others hitherto known. The most remarkable feature of this species consists in the presence of irregular spicules at the oscular margin.

Locality and Register No. of Specimen.—Wagu, Miye Prefecture; A 16.

16. *Leucandra fragilis*, n. sp.

(Pl. IV, fig. 6; Textfig. 8)

This species is represented in the collection by a single specimen (Spec. No. A 17) which was obtained at Wagu, Miye Prefecture.

It represents a flattened mass of approximately square shape, measuring about 15 mm. long and 10 mm. broad. The osculum at the upper end is slit-like and is naked.

The colour in alcohol is yellowish white and the texture is firm but brittle.

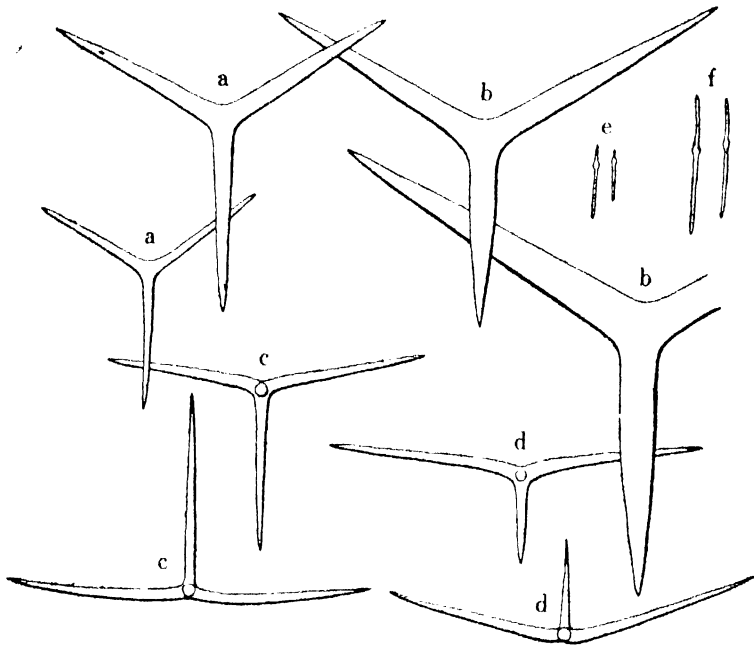
Structures.—The canal system is leuconoid. There is not a central gastral cavity, but branched narrow cavity runs irregularly interior of the sponge wall. The flagellate chambers are fairly thickly packed in the chamber layer. The exhalant canals are measured $300-570\mu$ in diameter.

The dermal skeleton consists of tangentially placed triradiates and of microxea in dense distribution. The skeleton of the chamber layer is made up of stout triradiates which are placed thickly and irregularly. The wall of the exhalant canals are sustained by quadriradiates with their apical rays projecting into the canal. The gastral skeleton is composed of tufts of microxea and of small tangential quadriradiates forming a few layers and with their apical rays projecting into the gastral cavity. The microxea project nearly vertically from the gastral surface and are grouped into small tufts around the apical rays of the gastral quadriradiates.

Spicules (Textfig. 8).—Dermal triradiates (a) regular or subregular.

Rays are straight, gradually and sharply pointed, $120\text{--}360\ \mu$ long and $16\text{--}36\ \mu$ thick at base.

Tubar triradiates (b) slightly sagittal. All rays are stout and straight, equally thick, sometimes slightly narrowed at base. Basal ray slightly shorter than paired rays, $230\text{--}640\ \mu$ long and $45\text{--}100\ \mu$ thick at base. Paired rays equal, tapering from base to sharp point, $270\text{--}700\ \mu$ long and $45\text{--}100\ \mu$ thick at base.



Textfig. 8. *Leucandra fragilis*, n. sp. a, Dermal triradiates. b, Tubar triradiates. c, Quadriradiates of larger exhalant canal. d, Gastral quadriradiates. e, Dermal microxea. f, Gastral microxea. (a-d $\times 100$, c, f $\times 200$)

Quadriradiates of exhalant canal (c) slender and sagittal. Basal ray straight, sharply pointed, $70\text{--}200\ \mu$ long and $8\text{--}16\ \mu$ thick at base. Paired rays equal, widely divergent, nearly equal in length with basal ray, $90\text{--}190\ \mu$ long and $8\text{--}16\ \mu$ thick at base. Apical ray straight, sharply pointed, slightly longer than facial rays, $100\text{--}210\ \mu$ long and $8\text{--}16\ \mu$ thick at base.

Gastral quadriradiates (d) sagittal. Basal ray straight, much shorter and slightly thinner than paired rays, $30\text{--}130\ \mu$ long and $8\text{--}15\ \mu$ thick at base. Paired rays widely divergent, sharply pointed, $90\text{--}320\ \mu$ long and $8\text{--}18\ \mu$ thick at base. Apical ray straight, thinner than facial rays, $100\text{--}130\ \mu$ long and $8\text{--}12\ \mu$ thick at base.

Dermal microxea (e) straight, sharply pointed at both ends. Close to one end occurs a nodiform ring. They measure $40-50\ \mu$ in length and about $2\ \mu$ in thickness.

Gastral microxea (f) nearly similar to dermal microxea in shape but longer than the latter, $70-90\ \mu$ long and about $5\ \mu$ thick at the ring.

Remarks.—This species bears a marked resemblance to the writer's *Leucandra dura*¹⁾ in many respects, but it may be easily distinguished from the latter by the differences in the microxea as well as by the external appearance.

Locality and Register No. of Specimen.—Wagu, Miye Prefecture, A 17.

17. *Leucandra abratsbo* HOZAWA

Leucandra abratsbo HOZAWA, 1929, p. 359, Pl. IX., figs. 57, 58; Textfig. 29.

In the collection there are twelve specimens of this species, all being obtained by the writer at Wajima. Each of them indicates a solitary individual with an elliptical osculum on its summit. They vary from 3 mm. to 10 mm. in length and are nearly similar in external appearance to one another, although some of them are laterally compressed or irregularly curved.

In respect to the canal system, skeletal arrangement and spiculation, these specimens are exactly identical with those of the type specimen.

Previously known Distribution.—Misaki Sagami Sea (HÓZAWA).

Locality and Register No. of Specimens. Wajima, Ishikawa Prefecture, B 5.

18. *Leucandra mediocanellata*, n. sp.

(Pl. IV, fig. 7; Textfig. 9)

The collection contains 27 specimens of this new species (Spec. No. C 5), which were obtained at Oshima, Miyagi Prefecture.

Each sponge represents a small solitary individual of ovoid shape showing on the upper surface a single circular osculum. The osculum is surrounded by a well-developed collar consisting of linear spicules. The dermal surface is highly hispid on account of the projecting oxea. The gastral surface appears slightly hispid from the apical rays of gastral quadriradiates. The colour in alcohol is yellowish white, and the texture is soft but elastic.

¹⁾ *Leucandra dura*, HOZAWA, 1929, pp. 371-373, Pl. XI., figs. 66-68; Textfig. 33.

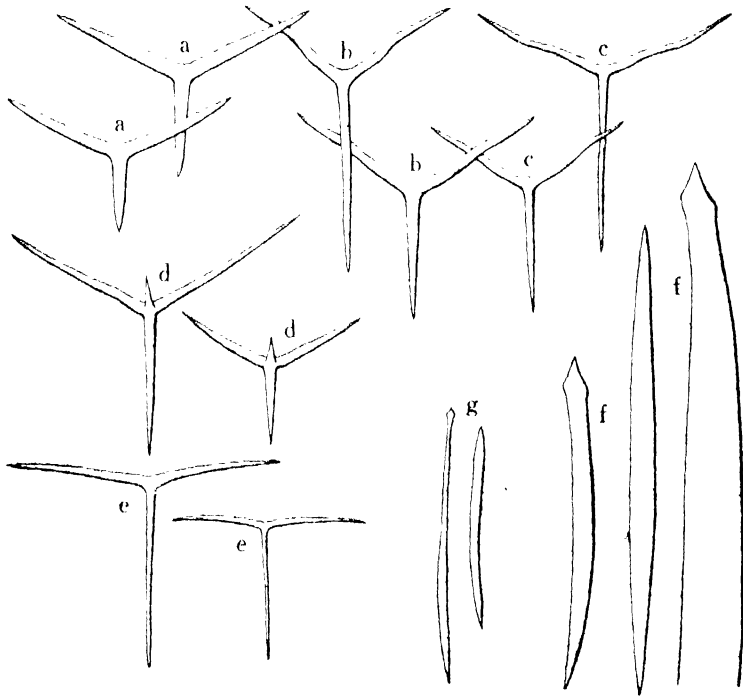
The largest specimen (Pl. V, fig. 11) upon which the further description is based is of an oval shape with a height of 6 mm. and a breadth of 2.5 mm. The collar of the osculum is about 2.5 mm. high. The gastral cavity is nearly straight and passes through the entire length of the body. It is circular in cross-section with a diameter of about 0.8 mm. The sponge wall is about 0.8 mm. in the thickest parts.

Structures.—The canal system is leuconoid. The flagellate chambers are spherical or oval in shape, being measured $60-80\mu$ in diameter.

The dermal skeleton is composed of tangential triradiates arranged in a few layers. The large and small oxea which occur in nearly vertical disposition in the sponge wall project outward beyond the dermal surface.

The skeleton of the chamber layer consists chiefly of triradiates irregularly arranged. Along the larger exhalant canals there occur some quadriradiates with their apical rays projecting into the canal.

The gastral skeleton is thin, consisting of triradiates and quadriradiates both of which being placed tangentially. The apical rays of the gastral



Textfig. 9. *Leucandra mediocanellata*, n. sp. a, Dermal triradiates. b, Tubar triradiates. c, Gasteral triradiates. d, Gasteral quadriradiates. e, Triradiates of oscular margin. f, Large oxea. g, Microxea. (a-f $\times 100$, g $\times 200$)

quadriradiates project into the gastral cavity.

The skeleton of the oscular margin is made up of slender oxea and of triradiates, all placed densely. The linear spicules run longitudinally and parallel with one another. The triradiates have their basal rays directed regularly downwards.

Spicules (Textfig. 9).—Dermal triradiates (a) sagittal and all rays are equal in thickness. Basal ray straight, shorter than paired rays, 45–150 μ long and 12–18 μ thick at base. Paired rays equal, straight or slightly curved, sharply pointed, 110–170 μ long and 12–18 μ thick at base.

Tubar triradiates (b) sagittal. Basal ray straight, usually longer than paired rays but sometimes equally long with the latter, 100–200 μ long and 14–18 μ thick at base. Paired rays nearly equal, doubly curved, forwards in the basal parts and backwards in the remaining parts, 110–170 μ long and 14–18 μ thick at base.

Gastral triradiates (c) sagittal and slender. All rays of equal thickness being 8–10 μ thick at base. Basal ray straight, slightly longer than paired rays, 150–250 μ long. Paired rays nearly equal, doubly curved as in the case of tubar triradiates and 120–190 μ in length.

Gastral quadriradiates (d) slightly sagittal. Basal ray straight, sharply pointed, shorter than paired rays, 90–160 μ long and 8–12 μ thick at base. Paired rays equal, straight or slightly curved, 110–220 μ long and 8–12 μ thick at base. Apical ray short, slightly curved, 40–50 μ long and 8–12 μ thick at base.

Triradiates of the oscular margin (e) sagittal. Basal ray straight, longer and thinner than paired rays, 150–190 μ long and about 8 μ thick at base. Paired rays equal, widely divergent, 120–150 μ long and 12 μ thick at base.

The large oxea (f) straight or slightly curved. Sometimes one end forms a lance-head, but some oxea are sharply pointed at both ends. They measure 450–2100 μ in length and 30–70 μ in thickness in the thickest parts.

Microxea (g) spindle shaped, slightly curved, broader nearer sharply pointed proximal end than the distal and which sometimes forms a lance head, about 180 μ long and 5 μ thick in the thickest parts.

Linear spicules of the oscular margin straight, slender, uniformly thick throughout their entire length and tapering towards both ends, which are finely pointed. They reach 2.5 mm. in length and are 6–10 μ in thickness.

Remarks.—In external form this species closely resembles *Leucandra spinosa* described above, but it may be easily distinguished from the latter by the difference in the arrangement of the skeleton and in the spiculation.

Locality and Register No. of Specimen.— Oshima, Miyagi Prefecture, C 5.

LITERATURE CITED

- BREITFUSS, L. (1897). Katalog der Calcareo der zoologischen Sammlung des königlichen Museums für Naturkunde zu Berlin. Arch. für Naturgesch., Jahrgang LXIII. Bd. I, pp. 205-226.
- (1898). Kalkschwammfauna der Westküste Portugals. Zool. Jahrb. Syst. Abt. Bd. XI, pp. 89-102, Taf. XI.
- (1932). Die Kalkschwammfauna der arktischen Gebietes. Fauna arctica, Bd. VI, pp. 237-252.
- (1935). Le Spugne calcaree dell' Adriatico con riflesso a tutto il Mediterraneo. R. comitato talassografico italiano, Mem. CCXXIII, pp. 1-15.
- BRØNSTED, H. V. (1926). Sponges from New Zealand. Part II. Papers from Dr. TH. MOITENSEN's Pacific Expedition 1914-16, XXXV. Vidensk. Medd. fra Dansk naturh. Foren, Bd. 81.
- (1932). Die Kalkschwämme der deutschen Südpolar-Expedition 1901-03. "Deutsche Südpolar Expedition" XX, pp. 1-47.
- CARTER, H. J. (1883). Further Observations on the So-called 'Farringdon Sponges', Calci-spongiae, ZITTEL followed by a Description of an Existing Species of a Like Kind *Leucetta clathrata*, new sp. . Ann. Mag. Nat. Hist., (Ser. 5), Vol. XI, pp. 20-37.
- (1886). Descriptions of Sponges from the Neighbourhood of Port Phillip Heads, South Australia. Ann. Mag. Nat. Hist., (Ser. 5), Vol. XVII, pp. 502-516.
- DENDY, A. (1891). Monograph of Victorian Sponges. Part I. The Organization and Classification of the Calcareo Homocoela, with Descriptions of the Victorian Species. Trans. Roy. Soc. Vict., Vol. III, No. 1, pp. 1-82, Pl. 1-9.
- (1892). Preliminary Account of *Synute pulchella*, a New Genus and Species of Calcareous Sponges. Proc. Roy. Soc. Vict., (n. s.), Vol. IV, pp. 1-6.
- (1893). Synopsis of the Australian Calcareo Heterocoela with a Proposed Classification of the Group and Descriptions of Some New Genera and Species. Proc. Roy. Soc. Vict., (n. s.), Vol. V, pp. 69-116.
- (1913). Report on the Calcareous Sponges collected by H. M. S. "Sealark" in the Indian Ocean. Trans. Linn. Soc. London, Zool. Vol. XVI, Part I, pp. 1-29, Pl. I-V.
- DENDY, A. and ROW, W. H. (1913). The Classification and Phylogeny of the Calcareous Sponges, with a Reference List of all the Described Species, Systematically Arranged. Proc. Zool. Soc. London, 1913, pp. 704-813.
- HAECKEL, E. (1870). Prodrömus eines Systems der Kalkschwämme. Jenaische Zeitschr., Vol. V, pp. 236-254.
- (1872). Die Kalkschwämme, eine Monographie. Berlin.
- HÔZAWA, S. (1916). On some Japanese Calcareous Sponges belonging to the Family Heteropiidae. Journ. Coll. Sci. Imp. Univ. Tokyo, Vol. XXXVIII, Art. 5, Pl. I-II.
- (1918). Report on the Calcareous Sponges collected by the U. S. Fisheries Steamer "Albatross" in the Northwestern Pacific during 1906. Proc. U. S. Nat. Mus., Vol. 54, pp. 525-556, Pl. 84-85.

- HOZAWA, S. (1928). Report of the Biological Survey of Mutsu Bay. 6, Calcareous of Mutsu Bay. Sci. Rep. Tôhoku Imp. Univ. Biol., Vol. III, pp. 219-221, Pl. I.
- (1929). Studies on the Calcareous Sponges of Japan. Journ. Fac. Sci. Imp. Univ. of Tokyo, Sect. IV, Zoology, Vol. I, Part 5.
- (1933). Report on the Calcareous Sponges obtained by the Survey of the Continental Shelf Bordering on Japan. Sci. Rep. Tôhoku Imp. Univ. Biol., Vol. VIII, pp. 1-20, Pl. I.
- JENKIN, C. F. (1908). The Marine Fauna of Zanzibar and British East Africa from Collections made by CYRIL CROSSLAND, M. A. in the Years 1901 & 1902. The Calcareous Sponges. Proc. Zool. Soc., London, Vol. II, pp. 434-456.
- KIRK, H. B. (1895). New Zealand Sponges. Third Paper. Trans. New Zealand Instit., Vol. XXVIII, pp. 205-210, Pl. III-IV.
- (1897). Notes on New Zealand Sponges. Fourth Paper. Trans. New Zealand Instit., Vol. XXX, pp. 313-316, Pl. XXXI-XXXII.
- LENDENFELD, R. von (1885). A Monograph of the Australian Sponges. Part III. The Calcispongiae. Proc. Linn. Soc. New South Wales, Vol. IX, pp. 1083-1150, Pl. LIX-LXVII.
- (1891). Das System der Kalkschwämme (Vorl. Mitth.) Sitzungsbericht Kais. Akad. Wiss. Wien. Mathem.-natur. Classe Bd. C. Abth. I, pp. 1-16.
- MINCHIN, E. A. (1896). Suggestions for a Natural Classification of the Asconidae. Ann. Mag. Nat. Hist., (Ser. 6), Vol. XVIII, pp. 349-362.
- ROW, W. H. and HOZAWA, S. (1931). Report on the Calcareous obtained by the Hamburg South-West Australian Expedition of 1905. Sci. Rep. Tôhoku Imp. Univ. Biol., Vol. VI, pp. 727-809, Pl. XIX-XXI.
- SCHMIDT, O. (1862). Spongien des Adriatischen Meeres. Leipzig, 1862.
- TANITA, S. (1939). Two New Calcareous obtained from Sasebo, Japan. Sci. Rep. Tôhoku Imp. Univ., Biol., Vol. XIV, pp. 319-326.
- TOPSENT, E. (1936). Étude sur des *Leucosolenia*. Bull. de L'Institut Océano., No. 711, pp. 1-47.
- URBAN, F. (1905). Kalifornische Kalkschwämme. Archiv für Naturgesch., Jahrgang. 72, Bd. I, pp. 33-76, Taf. VI-IX.
- (1909). Die Calcareous. Wissensch. Ergeb. der deutschen Tiefsee-Expedition (Voldivia), Bd. XIX, pp. 1-39, Pl. I-VI.

EXPLANATION OF PLATES

PLATE IV

- Fig. 1. *Leucosolenia ventosa*, n. sp. about $\times 2$.
Fig. 2. *Grantessa ampullae*, n. sp. about $\times 2$.
Fig. 3. *Leucandra rigida*, n. sp. about $\times 2$.
Fig. 4. *Leucandra spinosa*, n. sp. about $\times 2$.
Fig. 5. *Leucandra glabra*, n. sp. natural size.
Fig. 6. *Leucandra fragilis*, n. sp. about $\times 2$.
Fig. 7. *Leucandra mediocanellata*, n. sp. about $\times 2$.

PLATE V

- Fig. 8. *Paragrantia waguensis*, n. gen. and sp. about $\times 2$.
Fig. 9. The same. Part of horizontal section, showing the skeleton of apopyle (Ap). $\times 25$.
Fig. 10. The same. Skeleton of apopyle (Ap). $\times 200$.
Fig. 11. The same. Quadriradiates of apopyle. $\times 250$.



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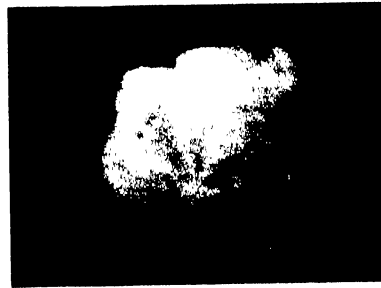
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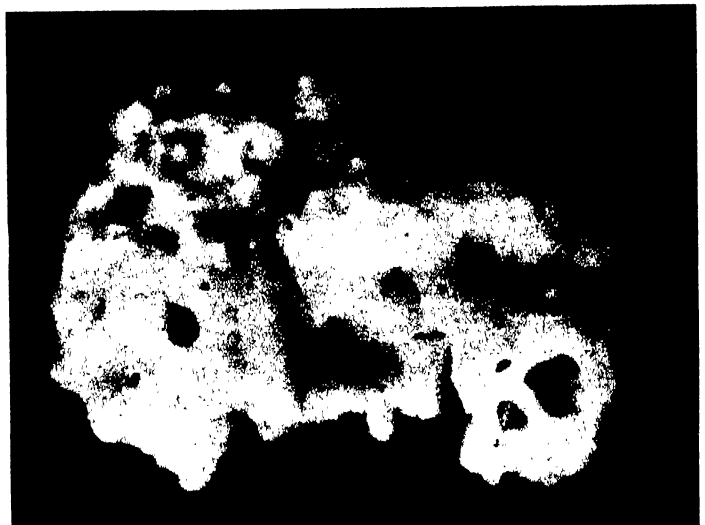
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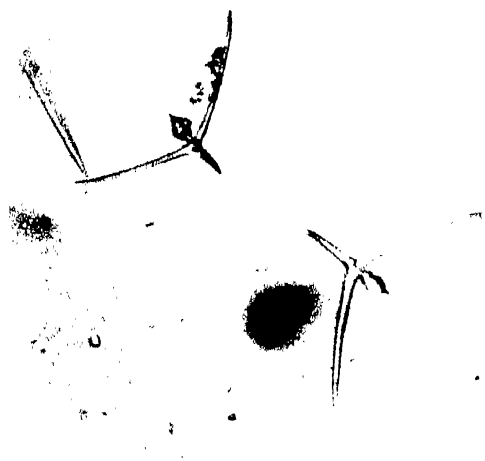


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THE HOMING, SPAWNING AND OTHER HABITS OF A LIMPET, *SIPHONARIA JAPONICA* DONOVAN*

By

NOBORU ABE

Institute of Biology, Kirin University, Kirin, Manchoukuo

(With 9 text-figures)

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INTRODUCTION

Siphonaria japonica DONOVAN is a limpet belonging to the Pulmonata, and is commonly seen on the rocky shore in the neighbourhood of Asamushi. The present writer has already studied another limpet, *Acmaea dorsuosa* GOULD=*Patelloida grata* (GOULD) (ABE, 1931, '32, '33), which is also commonly found in the same neighbourhood. He has also made observations of *Siphonaria atra* QUOY et GAIMARD, *S. sipho* (SOWERBY) and *Siphonaria* sp. (ABE, 1935, a, b, '39, a) which inhabit the neighbourhood of Palao Island, South Sea Islands. It is known that each of these limpets has its own home and has homing instincts at certain periods, as is also the case with limpets found in European seas which has been mentioned by many writers. The object of the present writer was to examine *Siphonaria japonica* in order to compare its habits with those of the other species. He specially wished to compare the habits of *Siphonaria japonica* living in the sea at 40° 55'-North latitude, with those of *S. atra* living in the tropical sea at 7° 20'-North latitude.

Both species belong to the same genus and their habitat is equally on rocky shores in littoral zones, but their environmental conditions, for instance, temperature, wave actions etc., are clearly different, and a comparison of the habits of the two species presented itself as highly interesting study. The observations which are here described were made chiefly in the year 1938 during the season from the spring onwards at the Asamushi Marine Biological Station facing Mutsu Bay.

Before going farther the writer wishes to express his sincere thanks to Prof. Dr. SANJI HÔZAWA, the Director of the Station, for the kind revision and correction of the present paper, and also his gratitude to

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the members of the same Station for the kind assistance given him in many ways.

I. HABITAT.

1. Horizontal distribution.

Siphonaria japonica DONOVAN inhabits mainly the rocky shore in the neighbourhood of the Asamushi Marine Biological Station, and it is also found attached to the piles of wooden bridges etc., as in the case of a bridge constructed in front of the Hotel Tookan, Asamushi. This species is not found on pebbly or on sandy shores.

But the *Siphonaria* is not found on the rocky northwestern shore of

TABLE 1. Vertical distribution of *Siphonaria* and the other inhabitants on the rock-face at Station 1 in the month of May. (May 14, 1938)

Species	<i>Siphonaria japonica</i> DONOVAN	egg-ribbons " <i>Patelloida pygmaea</i> DUNKER	<i>Cellana toreuma</i> REEVE	<i>Littorina littorita</i> <i>brevicula</i> PHILIPPI	<i>Chthamalus challengeri</i> HOEK	other animals	<i>Mitella mitella</i> LINNAEUS	<i>Porphyra</i> sp. 1.	<i>Ulva pertusa</i> KJELLM.	<i>Nemalion lubricum</i> DUBY	<i>Polysiphonia</i> sp.
Height in metres											
1.3-1.4				7							
1.2-1.3				3							
1.1-1.2				11							
1.0-1.1				0							
0.9-1.0				5							
0.8-0.9				49							
0.7-0.8			4	74			4				
0.6-0.7	1		4	93			5	2		5	
0.5-0.6	6		12	35	3/5		3	2	7	15	
0.4-0.5	6	1	2		5/5	c 1	3	12	50	41	
0.3-0.4	8	10		1	1/5	s 1/5		17		5	
0.2-0.3		1		5		s 5/5			12		1/5
0.1-0.2						s 1/5					2/5
0.0-0.1						c 1 b 2					S

c = Chiton, s = a Serpulid with calcareous tube of about 4 cm long, S = *Sargassum Thumbergii* (KUNTZ) OKAM. 3/5, means that it covers about 3/5 part of all the area in one section.

Gomi-jima, an island situated near the Station, but is found on its eastern shore. It was found very commonly on the western shore of the Station, but not on its northern shore when observed in 1938.

2. Vertical distribution of the *Siphonaria* and its relation to other organisms.

The writer has measured the number of various inhabitants found on a rock face standing nearly vertically, and have compared their zonations with the zone of *Siphonaria japonica*.

(i) Zonations in the months of May and June.

Station 1:

Station 1 is located on the face of a rock standing on the beach in the north of the Station. The waves beat vigorously on this rock-face in a strong west wind. The writer has measured the number of inhabitants found on the rock-face by means of the frame method during the time of low-tide. The breadth of the area measured is 20 cms, and the results thus far obtained are shown in Table I.

In Table I, the level of 50 cm is about the mean-tide-level, therefore the *Siphonaria* is distributed between the levels of about 10 cm over and 20 cm below the mean-tide-level, which about the same zone in spring and early summer as that of an alga *Porphylla*. It is commonly observed that the *Siphonaria* is hiding underneath the alga which is perfectly dried by direct sunlight in the time of low-tide. A view of Station 1 is shown on Fig. 1.



Fig. 1. *Siphonaria japonica* DONOVAN on the vertical rock face. Showing their egg-ribbons and the mating individuals (a). photo 7h 50m, a.m., May 27th, 1938.

Station 2:

Station 2 is on a rock-face at the north-western end of Hadakajima, a small island lying very close to the Station. This rock-faces westwards and the waves beat more vigorously upon it than upon the rock at Station 1 in both west and north winds. The writer has measured the inhabitants found on the rock-face by the frame method at low-tide, the breadth of measurement is 30 cms and the results thus obtained are shown in Table II.

TABLE II. Vertical distribution of *Siphonaria* and the other inhabitants on the rock-face at Station 2 in the month of June, (June 17th, 1938)

Height in metres	Species	<i>Siphonaria japonica</i> DOKOVAN and egg-tadpoles	<i>Patelloda pygmaea</i> DUNKER	other limpets	<i>Littorina Littorinaea</i> <i>brevicula</i> PUMPER	<i>Littorina Littorinaea</i> <i>millicera</i> PUMPER	bivalves	<i>Chthamalus chthamerei</i> HOLK	other animals	cellularous algae	<i>Chorieria clausenoides</i> HAYK. OKAM.	other algae
2.8-3.0					18	21		140				
2.6-2.8					22	34		140				
2.4-2.6					58	18		15				
2.2-2.4				P 20	92	18		12				
2.0-2.2					43	71		15				
1.8-2.0			1	P 108	12	10		13				
1.6-1.8			9		78	54		15				
1.4-1.6			1		2	1		55				
1.2-1.4			28		12			15	M 3			
1.0-1.2	2-3	20	C 2				S 11	15	M 1			p 1
0.8-1.0	2-2	8	C 6				My 33	14		13		g 3
0.6-0.8	2						My 51			45		g 7
0.4-0.6							My 36			45	5	b 1
0.2-0.4							My 6			45	3	
0.0-0.2										45		

P *Patelloda grata* GOULD, C *Cellana cucosmia* PHIBSBY, S *Septifer virgatus* WIEGEMANN, My *Mytilus crassitesta* (JASCHKE), M *Mitella mitella* (LINNAEUS), g green algae, b brown algae. * many hydroids, such as *Campanularia* etc., are attached to the shell of *Mytilus*

In Table II, the level of 0.8 m is about the mean-tide-level, therefore the *Siphonaria* is distributed between the levels of 10 cm over and 20 cm

below the mean-tide-level at the station. And it is noticed that the number of *Siphonaria* is clearly smaller than that seen in Station 1. *Patelloida grata* (GOULD) forms a colony at a higher level than the high-tide-level. *Cellana eucosmia* (PILSBRY) inhabits about the same level as that of *Siphonaria*.

When the data shown in Table I are compared with those given in Table II, it will be noticed that the zonations of animals and plants are different from each other in the two stations. And even in the same species of *Siphonaria*, the levels of the zonation are clearly different, namely, the zone of the *Siphonaria* at Station 2 is higher and broader than that in Station 1. And such differences of animals seems to be produced by the influence of wave actions, as the waves are always

TABLE III. Vertical distribution of *Siphonaria* and of other inhabitants found on the rock-face at Station 1 in the month of January. (Jan. 18th, 1939)

Species Height in metres	<i>Siphonaria japonica</i> DONOVAN	<i>Patelloida pygmaea</i> (DUNKER)	<i>Cellana toreuma</i> (REEVE)	<i>Littorina (Littorina) brevicula</i> (PHILIPPI)	<i>Chthamalus challengeri</i> HOEK	serpulid	other animals	<i>Mitella mitella</i> LINNAEUS	<i>Perophora</i> sp. 2.	<i>Polysiphonia</i> sp.
1.3-1.4				1					3	
1.2-1.3									15	
1.1-1.2				3					6	
1.0-1.1					1/10				25	
0.9-1.0		1			1/3				68	
0.8-0.9				6	4/5				11	
0.7-0.8		1		10	5/5			4		
0.6-0.7		7		46	4/5			5		
0.5-0.6	3	6		4	3/5			3		
0.4-0.5	2	2		1	1/3		c 1	3		
0.3-0.4	1				1/10	1/5				1/10
0.2-0.3	3	1	3			5/5				1/3
0.1-0.2						1/5	St 1, T 5			1/4
0.0-0.1							T 56			

c = Chiton, St = *Strongylocentrotus pulcherrimus* (A. AGASSIZ), T = *Thais* (*Mancinella*) *tumulosa clavigera* (KÜSTER).

higher and the water beats more strongly on the rock-face at Station 2 than at Station 1. The same phenomenon is also clearly seen in the zonation of a barnacle, *Chthamalus challengerii* HOEK.

(ii) Zonations in the month of January.

The writer has observed the inhabitants distributed on the rock-face of Station 1 in the winter time, in order to know whether the *Siphonaria* change their zonations or not during that time, and the results obtained are shown in Table III.

In Table III, the level of 0.5 metre is about the mean-tide-level, and the *Siphonaria* is distributed between the levels of about 10 cm over and 30 cm below the mean-tide-level, therefore the *Siphonaria* migrates to a zone about 10 cm lower in the winter time than in the months of May and June. And it is noticeable that many snails, *Thais* (*Mancinella*) *tumulosa clavigera* (KÜSTER), aggregate on the lowest part of the rock-face, though these snails are not seen there in the months of May and June, nor in the summer time. With regard to the zonations of algae, *Ulva* and *Nemalion* are not found in the winter time and *Porphyra* is found both in May and January, but the levels of their zones are quite different, and thus both the *Porphyra* seem to be of different species.

II. LOCOMOTION.

1. The times of the locomotion of *Siphonaria* and other animals.

The writer has observed the habitat of the *Siphonaria* at many times, at high-tide, mid-tide and low-tide, with the purpose of ascertaining the feeding-times of this animal. He was able to observe the movements of this limpet continuously on July 28th 1938 from time of high-tide in the morning till the rising tide was again at its highest in the afternoon.

As a consequence of these observations he is able to establish the fact that it is not the general habit of the *Siphonaria* to move about in the water, but that it only does so at stated times.

The results thus far obtained are shown in the following lines and the relation of locomotion of littoral animals to the tide-level is shown in Fig. 2.

July, 28, 1938.

Tide	{ High-tide: 3h 30m (0.8m), 16h 15m (0.8m)
	{ Low-tide: 10h 10m (0.1m), 22h 30m (0.3m)

7h 8m: The tide is ebbing and the water level is at about 20 cm above the habitat of *Siphonaria japonica* DONOVAN. All the *Siphonaria*

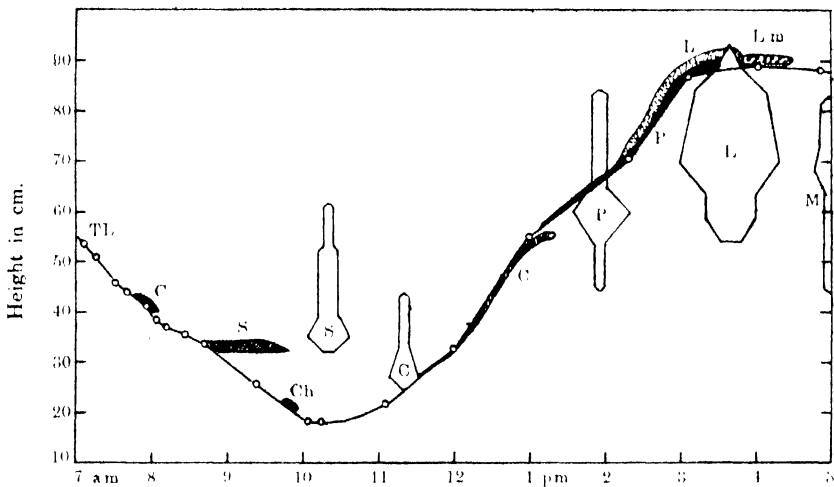


Fig. 2. Showing the times of the locomotion of *Siphonaria* and of other animals which occur in relation to the change of tide level. Polygonal areas show the vertical distribution of animals in the low-tide, and the area hatched or dotted show the locomotion of these animals following the movement of tide level. C, *Cellana toreuma* (REEVE); S, *Siphonaria japonica* DONOVAN; Ch, Chiton; P, *Patelloida pygmaea* (DUNKER); L, *Littorina littorivaga* brevicula PHILIPPI; Lm, *L. littorivaga* milligrana (PHILIPPI); M, *Mitella mitella* LINNAEUS. TL, tide-level.

were resting under the water on the rock and no movement was observed.

7h 38m: The tide level is lowering and is at the upper level of *Cellana toreuma* (REEVE) zone, and the *Cellana* begin a downward creeping slowly following the decrease of the tide level.

8h 3m: The *Cellana* have almost ceased movement, and the other individuals of the same species which are now just exposed to the air begin to creep downwards. Several young individuals of *Ligia exotica* (ROUX) of about 6 or 7 mm body-length also migrated downwards following the lowering of the tide-level, but the larger adult individuals did not migrate to the lower level but remained feeding at the high level near the high-tide-level.

8h 26m: The tide level is now about 1 cm above the *Siphonaria* habitat, and some of them begin to creep in the water, but a large number begin their creeping movement later just after they are exposed to the air.

9h 53m: Most of the *Siphonaria* have now returned and settled in their homes, and do not show any further locomotion. A chiton begins to creep in the area exposed to the air.

10h 6m: The tide reaches the lowest level, being about 15 cm lower than the *Siphonaria* habitat of this day. The chiton was resting within the crevices of a rock.

11h 0m: The tide begins to rise. The remaining *Siphonaria* who are still moving about return to their homes.

12h 0m: The tide rises and reaches the level of the home of the *Siphonaria*, but they remain fixed.

1h 0m: The tide rises and reaches the level of about 20 cm higher than the level of the home of *Siphonaria*, but the limpets are still resting. *Cellana toreuma* (REEVE) was creeping upward in the water following the rise of water level.

2h 20m: The tide rises to the level of about 45 cm higher than the *Siphonaria* habitat, but the species do not move. *Cellana* was hiding behind a rock.

3h 10m: The tide reaches the level of 50 cm higher than the *Siphonaria* habitat. A small limpet of another species, *Patelloida pygmaea* (DUNKER), was creeping upwards as if irritated by the rise of the water. A small individual of sea-hair, *Aplysia punctata* CUVIER, was creeping within the zone of *Chthamalus challengerii* HOEK.

4h 5m: The tide reaches its highest level of that day, and the lower part of the *Littorina millegrana* (PHILIPPI) habitat is washed by the waves, and the *Littorina* were creeping exposed to the air and no individual of the same was seen in the water.

4h 50m: The tide level is still at its highest. *Patelloida pygmaea* and *Littorina millegrana* were creeping just above the tide level. *Cellana toreuma* appeared on the front side of the rock creeping in the water. *Siphonaria japonica* which is now 55–60 cm below water level, is not moving.

From the above observations, the following facts have become clear; that the *Siphonaria* creeps just before or after the time when it is exposed to the air when the tide is falling, and that it continues to creep until the tide rises again and reaches the level of its habitat, and also that, as a general rule, this animal does not creep in the water at any other time.

The interesting fact emerges that the feeding-times of littoral animals are different from one another according to their species. *Cellana toreuma* creeps a little when it is exposed to the air and when it stops the *Siphonaria* begins to creep, followed by the chiton. *Cellana toreuma* creeps mainly in the water during the time of the rising tide, but *Patel-*

luida pygmaea begins to creep at the time just before the tide reaches its highest level. The same phenomenon of the sequence of the feeding-periods was also seen among the littoral animals inhabiting the rocky shore at Palao, and it seems to be a suitable arrangement for littoral animals crowded within a small area, who thus avoid struggles in securing their food.

2. The food of the *Siphonaria*.

The creeping movements of littoral animals is mainly determined by their feeding habits. *Siphonaria japonica* creeps in general on rock surface covered with a kind of brown algae as is seen in Fig. 3 and feeds on it and on some sedentary diatoms. But sometimes the writer has observed the *Siphonaria* feeding on the young alga of the *Enteromorpha compressa* GREV. and also on the *Porphyra* of deep reddish purple colour.

Several Siphonarians were observed in the laboratory and were examined at the time of excretion. Three of the larger limpets excreted two days after the time of feeding, and the smaller limpets of 5 mm shell length excreted the next day. Furthermore the writer has examined the excrement under the microscope. It is in the form of a half ring and is greenish-brown in colour. The greater part of the excrement consists of digested particles of brown algae and the remainder partly of diatoms, mainly of Naviculoid and *Melocira*, though the amount of these Diatoms is different according to individuals. And it was also noticed that only a few of the Diatoms were digested the majority of them being still living, and able to move slowly in the excrement when it was diluted with seawater. The excrement also contained sand-grains. In one case, many *Amoeba*, cubic in form were found in the excrement. About 20 individuals of the *Amoeba* were aggregated in one part of the excrement and they soon began to creep.

One limpet excreted in the laboratory about 10 days after it was first caught, and in this case the Diatoms found within the excrement were mainly of Naviculoid and *Melocira* forms and it was also seen that most of them were still living. And also many crystals of nearly cubical form and of reddish-brown colour were found in the excrement.

Judging from the results of these observations, it may be said that the food of *Siphonaria* consists mainly of algae, and that the Diatoms are not so important to it as food, but on the other hand, it is conceivable that the limpet acts as one of the agencies to transport the Diatoms.

3. Homing habits.

According to these minute observations of *Siphonaria japonica*, it becomes clear that it has the same homing habits as the other species of *Siphonaria*.

a) Home.

When the *Siphonaria* creeps away from its resting place, a scar of oval form is seen clearly on the rock face covered with algae and diatoms. The size of the scar is the same as that of the shell aperture of the limpet as is shown in Fig. 3.

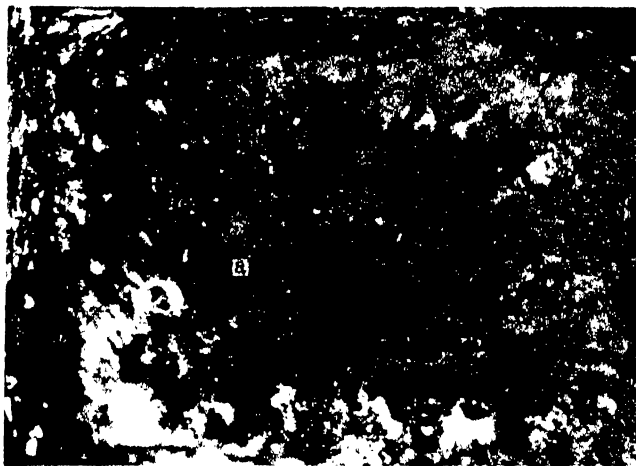


Fig. 3. Showing the home a of *Siphonaria japonica* DONOVAN
photo 11h a m., June 18th, 1938.

The home shown in Fig. 3 is that formed on a comparatively flat rock face, and there are also many other homes formed on a narrow area in the zone of the barnacle, *Chthamalus challengerii* HOEK. The homes shown in the latter position have no smooth margins of oval form, and the margins of the shells of the *Siphonaria* are also irregular and are quite fitted to the unevenness of the surroundings. Sometime individuals are found with their shells covered with short algae, and on this account it is rather difficult to distinguish the shells from their surroundings.

b) The duration in time and the paths of the feeding locomotion.

The duration in time and the paths of the feeding locomotion of *Siphonaria* were precisely examined in an intensive observation made on July 28th 1938 and a detailed record was taken of each individual as shown in the following lines and in Fig. 1.

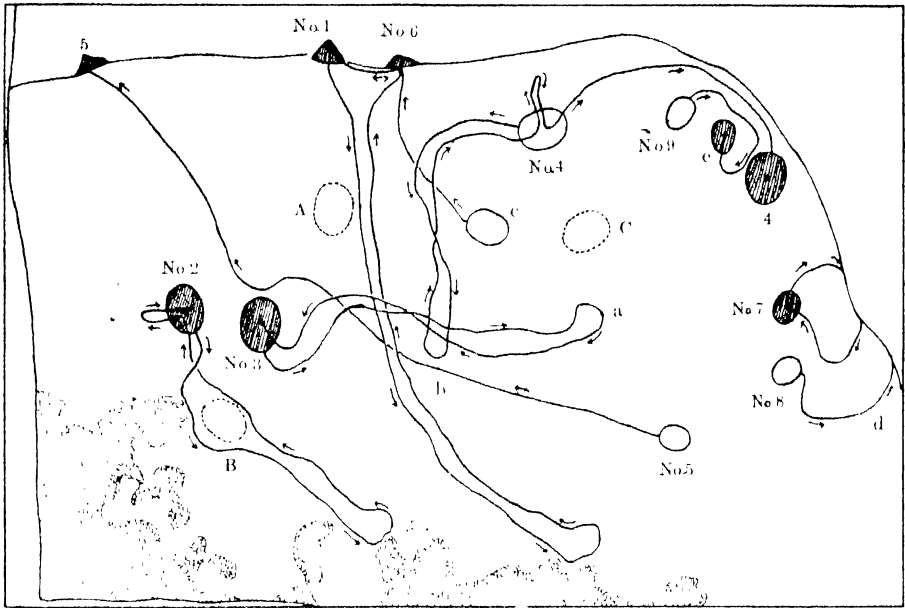


Fig. 4 Foraging paths of the *Siphonaria japonica* DONOVAN. Arrows indicate the direction of creeping.

Animal No. 1: Shell length 11.0 mm. At 8h 26m, the writer found a *Siphonaria* creeping in the water about 5 mm distant from its home, and it continued feeding in this position as is shown in Fig. 4, No. 1; and at about 8h 45m of the same day it turned creeping in the direction of its home, the path taken on the way home being very close to that taken on the way out. But at 9h 57m, it entered the home belonging to individual No. 6 (individual No. 6 was at that time absent not having yet returned). It did not stay here long however, but began to creep again after making a circular movement and it arrived back at its own home at 10h 1m. Coming back to its own home it made a circulating movement for several seconds and finally settled down. The total length of the paths going and returning was about 29.5 cm.

Animal No. 2: Shell length 10.5 mm. At 8h 39m, the tide having lowered the *Siphonaria* was exposed to the air for the first time. At 8h 41m, it began to creep on the exposed rock and continued feeding tracing food paths as shown in Fig. 4, No. 2. At 10h 5m, it returned to its home, and there it made a circulating movement. At 10h 13m, it began to creep out again and was seen feeding at a position 5 mm distant from its home, to which it soon after returned. At 10h 15m, it

remained settled in its home. Total length of paths followed by the limpet this time was about 7.0 cm.

Animal No. 3: Shell length 12.0 mm. At 8h 39m, it was exposed to the air for the first time; at 8h 40m, it began to move about in its home; at 8h 47m, it began to creep out; at 9h 10m, the limpet rested at its destination in the feeding ground as shown by (a) in Fig. 4. It stayed there about an hour; at 10h 31m, it was found creeping on its way home and at 11h 0m, it reached its home and remained there. Total length of paths thus followed by this limpet was about 6.5 cm.

Animal No. 4: Shell length 11.0 mm. At 8h 49m, it was exposed to the air. At 8h 51m, it began to creep out from its home and it traced a path seeking for its food as shown in Fig. 4, No. 4. At 9h 12m, it returned to its home and was seen making a circulating movement there. At 9h 13m, it again began to creep out from its home, but after a creep of only about 2 cm it returned again to its home at 9h 15m and remained there. At 9h 20m, it was found that the limpet was making a circulating movement in its home. It began to creep out once more from its home at 9h 48m. At 10h 13m, it entered home of individual No. 4' and remained there, not creeping any more.

Animal No. 5: Shell length 6.0 mm. At 9h 40m, it was found creeping at the point (b) shown in Fig. 4, and it was resting on the point No. 5 at about 9h 0m, but after that it continued to creep. At 10h 0m, it reached the position No. 5' and rested. The total length of the path was about 23.0 cm.

Animal No. 6: Shell length 9.5 mm. At 10h 17m, the limpet was found creeping at the point (c), and it appeared to be on its way home. At 10h 32m, it returned to its home, No. 6, and rested there. The distance from (c) to this home is about 7.5 cm, therefore the total length of its creeping may be estimated at about 15 cm.

Animal No. 7: Shell length about 6 mm. At 8h 34m, it was exposed to the air for the first time; at 8h 38m, it was found making a circulating movement in its home. At 8h 40m, it began to creep out. At 8h 58m, it returned to its home. Total length of the path was about 5 cm.

Animal No. 8: Shell length about 6 mm. At 8h 34m, it was exposed to the air; at 8h 38m, it was making a circulating movement in its home. At 8h 40m, it began to creep out. At 9h 16m, it was found creeping towards the lower part of the side of a rock.

Animal No. 9: Shell length about 6 mm. At 8h 49m, it was ex-

posed to the air ; at 8h 50m, it began to creep out from its home. At 9h 4m, though the limpet was on its way home, halted at (e) point shown in Fig. 4 and did not creep any further.

From the data given above, it seems to be clear that the direction of locomotion varies with the individual, but the great majority of these animals seem to creep towards a level higher than that where their homes are found at least they do so when they start creeping. The distance the limpets travel also varies with the individual. In general, the maximum limit of distance attained on the foraging paths seems to be within 15 cm from the home. And the distances reached by the limpets when feeding seem to be related to the volume of food captured. Thus some limpets begins to creep out again after they have returned to their homes. Animals No. 2 and No. 4 did so twice or three times to take more food. And it is noticeable that the second or third journeys were undertaken after the limpet had definitely completed its first journey and had returned back into its home. This fact seems to be important when considering the homing instincts of this kind of animal.

Six of the nine individuals above alluded to returned exactly to their homes but the remaining three did not do so, and it was noticed that the latter individuals were of small size in body with a shell of about 6 mm long. But even larger individuals with a shell of about 9-11 mm long have sometimes mistaken the way to their homes. For instance, home A, B and C shown in Fig. 4 remained empty on the day July 28th, though they had been found occupied by Siphonarias on the day June 16th.

It is an interesting fact that one limpet, viz. individual No. 5, had two homes and it used to creep from one home to the other. The nature of the homing instinct of this limpet seems to be rather different from that of the others, and it may be looked upon as an exceptional case.

III. REPRODUCTION.

Concerning the reproduction of *Siphonaria*, HUTTON (1882) studied the development of *Siphonaria australis* QUOY and GAIMARD, FUJITA (1895, 1904) made observations of the formation of germinal layers in the case of *Siphonaria lepida* GLD., and the present writer (ABE, 1939, a) dealt with the mating and spawning habits and the early development of *Siphonaria atra* QUOY et GAIMARD. But these habits and development in the case of *Siphonaria japonica* DONOVAN seem not yet to have been fully studied.

1. Mating habits.

Siphonaria japonica is hermaphrodite in nature as is the case with other species of Pulmonata. The manner of the mating of the *Siphonaria* is identical with that of *Siphonaria atra* QUOY et GAIMARD, namely, two individuals come very close the sides of their heads coming in contact as is seen in Fig. 1, a. The penis on one individual is extruded on the right side of the head and is intruded into the genital pouch of the other reciprocally. When the writer tried to separate the mating individuals, he found a large amount of white mucus secreted inside the genital pouch, and moreover, there was left a long thread-like process of white colour in the same pouch. The shape of this process is shown in Fig. 5, and it closely resembles the spermatophore of *Siphonaria australis* described by HUTTON (Ann. & Mag. Nat. Hist. S. 5, Vol. 9, Pl. XV, Fig. 7).

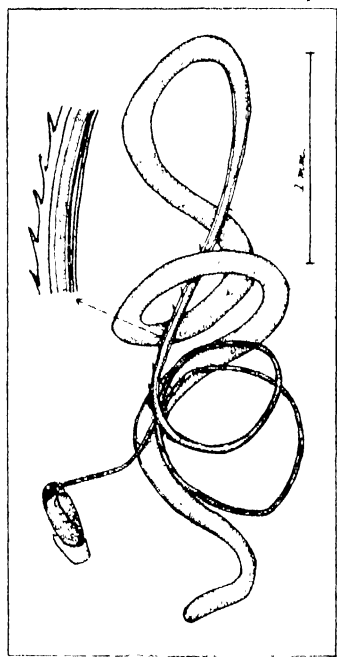


Fig. 5. Spermatophore of *Siphonaria japonica* DONOVAN.

The spermatophore of *Siphonaria japonica* is about 12.0 mm in length and contains numerous spermatozoa in the interior of its thicker part. The top of the spermatophore gradually narrows to form a filament which is hollow inside and provided with a number of minute spines on its outer surface as shown in Fig. 5, b.

2. Spawning habits.

The writer was able easily to observe the egg-ribbons laid on the littoral zone in the months of May, June and July, and sometimes he was also able to observe the *Siphonaria* in the act of spawning at the time of low-tide. In the act of spawning, the *Siphonaria* creeps out from its home and settles down at a place on the surface of some comparatively smooth, and then the egg-ribbon is emitted passing the siphonal groove and through the genital pore. Thereafter the limpet turns slowly counter-clock-wise, the egg-ribbon thus taking a semi-circular or nearly circular shape. The egg-ribbon laid thus is shown in Fig. 6, and others of varying shapes are also seen in Fig. 1.

The positions where the egg-ribbons are laid are generally just below the level of the home of *Siphonaria* as shown in Table I, II and Fig. 1. And it is noticeable that the egg-ribbons are laid in groups in several places. But the number of egg-ribbons in each group is different according to their position on the rock and also according to season. The number of egg-ribbons found in the different groups was respectively 5, 6, 11, 12, 12, 16, 17, 22, 24 and 29 when observed on May 28th 1938.



Fig. 6. An egg-ribbon of *Siphonaria japonica* Doxovax which was laid in the laboratory—photo 7h 30m, a.m., July 1st, 1931

The size of the egg-ribbon is nearly the same as that of the shell of the mother individual in most cases, but sometimes it is very small, though such small ones are usually only observed at the end of the spawning season, that is in July.

3. Egg and larva.

The writer secured half an egg-ribbon which was laid at 9h 20m a.m. on May 12, and tried to culture it in a glass vessel measuring 9 cm in diameter and 2 cm in depth. The sea-water contained in the vessel was changed once a day the water being taken from the neighbourhood of the habitat of the *Siphonaria*. The glass vessel was placed close to a glass window and the development of the eggs was observed.

a) Description of egg.

There are many eggs enclosed within the egg-capsule and they are arranged in several layers in the opaque gelatinous substance forming the egg-ribbon. The egg-capsule is elongated oval in shape with the longer diameter of about 273μ and the shorter diameter of about 154μ .

and both ends of the longer diameter are thickened. The egg is light cream-yellow in colour, and is spherical in form with the diameter ranging from 87 to 91 μ , and it is surrounded by a granulated opaque substance (Fig. 7, a).

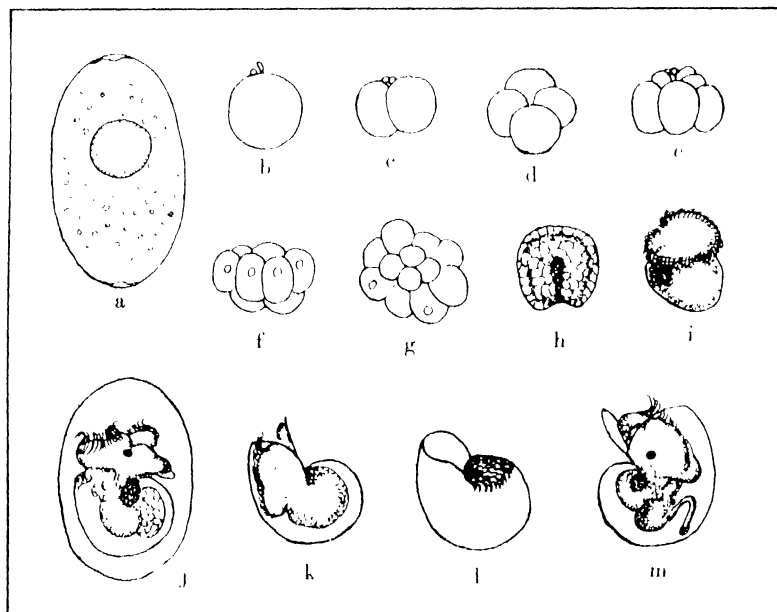


Fig. 7. Development of the egg of *Siphonaria japonica* DONOVAN (about $\times 108$). Of the explanation of figures a-l, see text. m: the larva of two days after hatching.

b) Development of egg.

The development of the eggs of the *Siphonaria lepida* GOULD is exhaustively studied by FUJITA (1904) and the development of the egg of *Siphonaria japonica* DONOVAN closely resembles this species.

The eggs which the writer placed in the glass dish were laid at 9h 20m on May 12th, but they did not show cell-division till 6h p.m. of the same day. (Water temperature was 14.0 C.)

At 7h 30m 15m a.m. May 13th. (Temp., 14.7 C.)

Eggs of various stages were found in the early cell division on May 12th, viz. the stage of expulsion of two polar bodies (Fig. 7, b), the stage of two cells of about equal size (Fig. 7, c), the stage of 4 cells (Fig. 7, d) and that of 8 cells (Fig. 7, e). Above all the stages of 8 cells predominated the arrangement of 8 cells in this stage closely resembling that seen in the case of *Siphonaria atra* QUOY et GAIMARD, namely 1

of these cells which may be micromeres are clearly smaller than the remaining 4 which may be looked upon as macromeres.

At 1h 50m-2h 20m p.m.: Most of the eggs were in the state of cleavage and began to form the second ectomere-quartets (Fig. 7, f).

At 7h 20m-50m a.m. May 14th. (Temp. 13.0°C): Most of the eggs reached the stage of second ectomere-quartet perfectly (Fig. 7, g).

On May 17th, (Temp. 15.0°C): The eggs developed into the blastula stage as shown in Fig. 7, h.

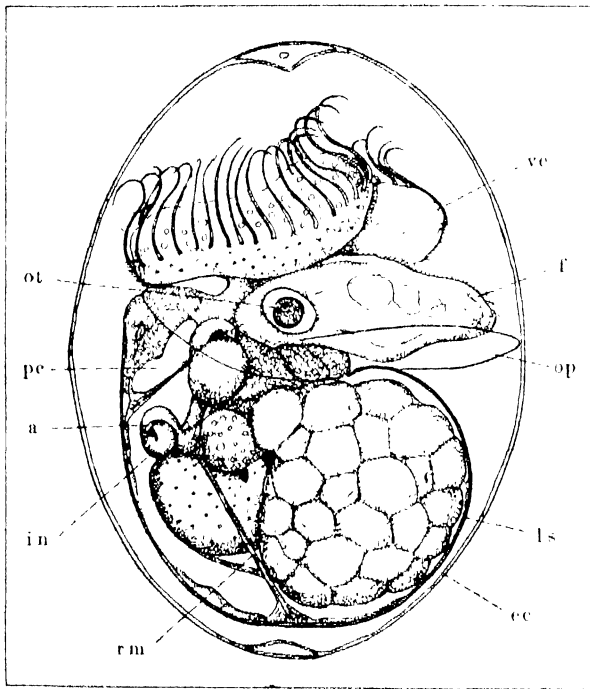


Fig. 8. Post-larva of *Siphonaria japonica* DONOVAN just before hatching. (about $\times 400$). ot., otocyst, pe., pallial cavity, a., anus, in., intestine, rm., retractor muscle, ve., velum, f., foot, op., operculum, ls., larval shell, ec., egg-capsule.

On May 20th, (Temp. 13.0°C): The eggs attained the early stage of veliger (Fig. 7, i). The velum was bilobed and the flagellae found on the margin of the velum were moving actively and were thus rotating inside the egg-capsule. The length of the flagellae above mentioned was from 8 to 9 μ . At this stage the larval-shell was not yet formed.

On May 26th, (Temp. 25.0°C): The larval-shell was well formed,

and the otocyst was clearly seen but it had not yet developed into the form of a double ring, having its diameter of about 8μ (Fig. 7, j).

On May 30th, (Temp. 15.8°C): The post larva of veliger was well developed as shown in Fig. 8. The otocyst became larger having developed into the form of a double ring, and the diameter measured about 11μ . The length of each flagellum found on the velum was about $55\text{--}57\mu$; and a number of short cilia of about 6 or 7μ long were all over the surface of the rudimentary foot and in the lower part of the velum, these were seen to produce a current running towards the mouth. The larva was still contained within the egg-capsule, and was rotating slowly. The operculum was moving rhythmically and slightly as if it were affected by the pulsation of heart, the number of rhythmical movements of the operculum was from 32 to 50 in 15 seconds.

On May 31st, (Temp. 16.2°C): A number of the larvae were already hatched out at 9h in the morning, and they were found on the bottom of the vessel showing a right-handed rotating movement. When the larva swims, it is seen that the shell is kept posteriorly. The shell of the larva just hatched was measured and was found to be about 0.16–0.18 mm in the longer diameter, and 0.11–0.115 mm in the shorter diameter, and about 0.095–0.10 mm in thickness (Fig. 7, k and l). The larval-shell is light cream-yellow in colour generally, and its apex is brown and some wrinkles are seen on it. The wrinkles are rougher than those found on the larval-shell of *Siphonaria atra* QUOY et GAIMARD.

4. The time of mating, spawning and hatching of larva.

An egg-ribbon of the *Siphonaria* laid on May 5th and found by the writer contained eggs which had already reached the gastrula stage in their development. On the other hand several individuals were found mating on May 7–12th, and the spawning was observed during the following days. Many more egg-ribbons were found on May 20th, but the mating had not been observed. On May 27th, the writer found one pair mating but could not find the individuals later in the act of spawning, though the egg-ribbons laid were very numerous, for instance about 140 egg-ribbons were found in the neighbourhood of Station 1 only. On June 10th, one pair of animals was found mating; and on June 15th, also one pair of animals mating, and a number of newly spawned egg-ribbons. On June 17th, and 19th many egg-ribbons laid and several spawning individuals were noticed; on June 20th, 2 pairs of animals were found mating. But on June 27th, most of the egg-ribbons were already hatched and no mating individuals were observed. On July 2nd,

one pair of mating individuals were seen, and egg-ribbons about to hatch. But on July 16th, some egg-ribbons were again found. On July 19th, some newly-laid egg-ribbons, about 11 in number, were found. On July 25th, the majority of egg-ribbons was hatched, and only a few remained unhatched. On July 27th, there were no egg-ribbons of the *Siphonaria* to be seen in its habitat, nor were there any on the following days, nor from 13th to 17th of August. No eggs were found from September to January, nor throughout the winter months, nor were they found in April 1939.

Judging from the above facts, we are convinced that there are periodicities of spawning and mating in the case of *Siphonaria*. In comparing these data with lunar periodicity, the writer have obtained the results as shown in Fig. 9.

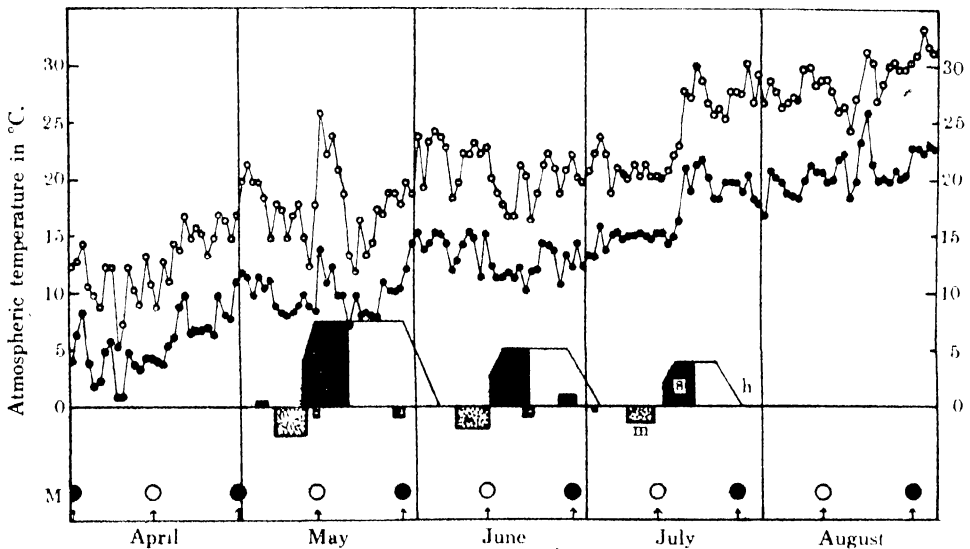


Fig. 9. Relation of the times of mating, spawning and hatching of the *Siphonaria* to lunar periodicity. m., mating, s., spawning, h., hatching, M., phase of moon. ○—○—○—maximum temperature, ●—●—●—minimum temperature. (Temperature of sea water is always between the both temperatures of maximum and minimum.)

From Fig. 9, it is clearly noticeable that the mating of the *Siphonaria* occurs or begins mainly at the time of the waxing half-moon, the spawning at the time of the full-moon, and the hatching of the eggs mainly at the time of the new moon.

The above facts seem to coincide well with the fact that the spawning

occurs about 7 days after mating. For example, a *Siphonaria* which mated on June 10th, 1938, was brought to the laboratory and cultured in a glass vessel containing a small piece of rock. And it laid an egg-ribbon on June 15th. Another *Siphonaria* which was found at Gomi-jima was brought to the laboratory, and after about 6 days (on July 1st, 1931) it laid an egg-ribbon from which the larvae hatched on July 15th.

The egg seems to take about 20 days to hatch when kept in the laboratory, which seems presumably a longer time than in its natural conditions. And it is also likely that the time spent in hatching is shorter in June and July than in May, on account of the increase of the temperature of the seawater. For instance, the temperature of the seawater was about 13°C on the days following the full-moon which occurred in May, while it was 17-18°C on the days following the full-moon in June, and 20-23°C in the days following the full-moon in July. The time taken in the hatching of the egg is well seen in Fig. 9.

But it is also noticeable that there are exceptions and that a few individuals of the *Siphonaria* will mate in the days of the waning half-moon as seen on May 27th, or in the days of the new moon as seen on July 2nd. Thus there are cases in which the time of spawning and of hatching may differ from the normal, but these are few in number. We may consider that the mating and spawning season of *Siphonaria japonica* DONOVAN is during the months of May, June and July, and that this season appears to be related to lunar periodicity.

IV. GENERAL CONSIDERATIONS.

1. On the level of habitat.

It is generally accepted that littoral animals are distributed in definite zones and that the zonation of each animal or plant is clearly limited. The zonation of animals has been diligently studied by many scientists in different countries, and the results of their researches are well represented when we mention works by FLATTERLY and WALTON (1922), PEARSE (1936), STEPHENSON (1936, '39), BRIGHT (1938), BOKENHAM, NEUGEBAUER and STEPHENSON (1938), EYRE (1939), LINKE (1939), etc. The zonation of animals inhabiting intertidal zones in tropical seas has also been studied by several investigators including the present writer ABE (1937).

He was also observed the zonation of some animals found in the district surrounding the Asamushi Marine Biological Station, studying the following species, *Acmaea dorsuosa* GOULD=*Patelloida* (*Tectura*) *grata*

dorsuosa (GOULD) (ABE, 1931, '33), *Littorina* (*Littorivaga*) *brevicula* (PHILIPPI) and *L. (Littorivaga) millegrana* (PHILIPPI) (ABE, 1935, c), and *Siphonaria japonica* DONOVAN, *Patelloida pygmaea* (DUNKER), *Cellana toreuma* (REEVE), *Cellana eucosmia* (PILSBRY) and sedentary animals, *Septifer virgatus* (WIEGMANN), *Mytilus crassitesta* LISCHKE, *Chthamalus challengerii* HOEK, etc.

The levels of the habitats of littoral animals are decided according to the duration of the periods of time in which the animals are exposed to the air, and submerged in the water. But the changes of temperature of seawater also influences the zonation of some littoral animals, and thus a seasonal migration is seen in the case of *Ilyanassa obsoleta*, *Littorina litorea*, *Littorina rudis* (BACHELDER, 1915, cited from LINKE, 1935), *Acmaea dorsuosa* GOULD (ABE, 1933), *Littorina* (*Littorivaga*) *brevicula* (PHILIPPI) (ABE, 1935, c), *Melanoides* (*Semisulcospira*) *niponicus* (SMITH) (MORI, 1937), etc.

From the observations made at Palao, it was noticed that the habitat of *Siphonaria atra* QUOY et GAIMARD lies between the levels of mean-tide and of 30 cm below mean-tide, and that the habitat of *Siphonaria siphon* SOWERBY lies at a level just above the zone of the former species (ABE, 1935, a, '39, a). In the neighbourhood of the Asamushi Station, *Siphonaria japonica* DONOVAN inhabits a zone in early summer between the levels of about 10 cm above and of about 30 cm below mean-tide-level at Station 1, and between the levels of about 40 cm above and of about 20 cm below mean-tide level at Station 2; while the same species is found between the levels of about 10 cm below and of about 60 cm below mean-tide-level in Station 1 in winter time.

Therefore, the breadth of the zones of both *Siphonaria atra* and *S. siphon* is narrower than that of *S. japonica*. When we compare the breadth of the zone of *S. atra* with that of the inter-tidal zone, the ratio is about 1/8, while that of *S. japonica* is about 1/2 even in the early summer. This difference in the breadth of the zones may be according to the difference of species, but it seems more likely to be caused by the difference of wave action in the two cases.

And it is also noticeable that the shifting of the zone level that occurs in the case of *S. japonica*, is presumable influenced by the decrease of temperature, though this is not so clear as in the case of *Acmaea dorsuosa* GOULD (ABE, 1933). Such seasonal shifting is not seen in either *Siphonaria atra* or *S. siphon*, these species living in tropical seas where the temperature of the seawater and of the air is almost constant throughout the year.

2. The times of locomotion for feeding purposes.

Many observations have been made on the times of locomotion of marine animals living in littoral zones, but it seems to the writer that they are not yet complete as it is difficult to observe the creatures in their natural surroundings. Even of one species, there are many different opinions as in the case of *Patella vulgata* (ORTON, 1929). RUSSELL (1934) says of this limpet that, "The limpets living near high-water mark do not move very much or very far from their home, and only when the sea covers them. Those living farther down the beach move about more freely — generally when they are submerged, but also when uncovered by the tide, if the air is moist."

In the seasons spring and summer, *Acmaea dorsuosa* GOULD which lives above the high-water mark does not creep for feeding except when the waves splash it (ABE, 1931), and even then only certain individuals in a colony do so.

Siphonaria atra QUOY et GAIMARD and *S. sipho* SOWERBY begin to creep just before or just after the exposure of the shell to the air during the ebbing-tide (ABE, 1935, a, '39, a). In the course of the present observations, it became clear that one species of limpet, the *Cellana toreuma* (REEVE), creeps mainly when the water is rising and just covering the limpet, while another species *Patelloida pygmaea* (DUNKER) creeps mainly in the open at the time just before the tide reaches its highest level, and *Siphonaria japonica* DONOVAN creeps just after it is exposed to the air during the ebbing-tide.

Among the species belonging to the Littorinidae, *Littorina exigua* DUNKER (MITSUKURI, 1901), *L. littorea* (BOHN, 1909; MORSE, '10; HASEMAN, '11), *L. rudis* (BOHN, 1905; MORSE, '10), *L. obtusata* (BOHN, 1905), *L. (Littorivaga) brevicula* (PHILIPPI) (ABE, 1935, c), etc. were studied formerly, and the writer has also made some observations of *L. (Littorivaga) millegrana* (PHILIPPI). It is known that all these species in their natural state exhibit rhythmical movements synchronous with the rise and fall of the ocean-tide. BOHN, 1905, stated that these rhythmical movements are seen even in the laboratory, but HASEMAN and ABE were not able to observe any such movements. *Melarhaphe (Littorinopsis) scabra* (LINNAEUS) is known to exhibit rhythmical movements synchronising with the tidal rhythm, though this species lives on the land being mainly found on mangrove-trees (ABE, 1936, '39, b).

Among the species belonging to the Neritidae, *Nerita japonica* DUNKER (TANITA, formerly SUZUKI, 1935) creeps as a rule when the tide rises and

reaches it, while *Nerita* (*Theliostyla*) *exuvia* LINNÉ and *Nerita* (*Ritena*) *undata striata* BURROW begin to creep during the ebbing-tide just before when their shells are exposed to the air (ABE, 1938).

Among the chitons, *Ischinochiton magdalensis* creeps only at night, while *Chiton tuberculatus* creeps mainly at high-tide, though it creeps also at low-tide to some extent in damp places (AREY and CROZIER, 1919). The other species of chiton, *Liolophura japonica* (LISCHKE) creeps during the ebbing-tide just before or just after the body is exposed to the air as mentioned in the present study.

Onchidium (*Onchidella*) *floridanum* DALL creeps at least an hour before the rising-tide reaches its home (AREY and CROZIER, 1918). *Onchidium* sp. which is commonly seen in the littoral regions of the Palao Island creeps during the ebbing-tide as soon as its body is exposed to the air (ABE, 1938).

As is stated above, the time of locomotion differs in various species even though they belong to the same genus. But it is clear that the locomotion of littoral marine animals is mainly in relation to the movements of the tide and of the waves. The changes of moisture-conditions which occur in consequence of the movements of the tide also act as an important factor in the locomotion of animals which creep mainly when exposed to the air, and this is seen specially clearly in the case of *Melarhaphe* (*Littorinopsis*) *scabra* (LINNAEUS) (ABE, 1936). On the other hand, light-intensity is an important factor in the case of animals which creep only in the night-time as *Ischinochiton magdalensis* (HEATH, 1939), *Cerithium* (*Aluco*) *aluco* (LINNÉ), *Chrysostoma paradoxum* (BORN) (ABE, 1937, b), etc. Light-intensity may also affect those creep only in the daytime as *Batillaria multiformis* LISCHKE (ABE, 1934), those which creep both in the day and in the night as in *Siphonaria atra* QUOY et GAIMARD (ABE, 1935, a, '39, a), and also those which creeps more actively in the night-time than in the daytime, as *Melarhaphe* (*Littorinopsis*) *scabra* (LINNAEUS) (ABE, 1936, '39, b). The combination of light-intensity with tidal-rhythm constitutes an important factor in some species when they begin to creep. And there exist several phenomena which seem to have some relation to the phases of the moon, for instance as seen in the increase of the number of migrating individuals in the case of *Batillaria multiformis* (ABE, 1934) and in the changes in the number of mating individuals in the case of *Melarhaphe* (*Littorinopsis*) *scabra* (ABE, 1936, '39, b), etc.

Therefore, it is recognised that the locomotion of littoral animals is

the result of complex phenomena influenced by many factors, and moreover when their locomotion is observed in a limited area, it is apparent that there exists a well-ordered sequence of locomotion for feeding purposes as is described in the present observations. The same phenomena were also observed at Palao, namely in the cases of zonations of *Littorina* (*Littorivaga*) *granularis* (GRAY), *Siphonaria siphon* SOWERBY, *S. atra* QUOY et GAIMARD, *Planaxis sulcatus* (BORN), *Cerithium concisum* var., etc. (ABE, 1937, b). The two kinds of *Siphonaria* above mentioned creep mainly during the ebbing-tide after they are exposed to the air (ABE, 1935, a, 1939, a), and *Planaxis* creeps during the ebbing-tide at a depth of about 50 to 60 cm (ABE, 1938), while *Cerithium* creeps at the time of high-tide. These two snails, *Planaxis* and *Cerithium* creep and climb up to the zone of *Siphonaria*, but their times of feeding are clearly different from that of the latter.

On comparing the habits of *Siphonaria japonica* with those of *S. atra*, it is found that both species begin to creep just before or just after the time when the body is exposed to the air, and the creeping continues from about 20 minutes to 2 hours in the former species and from 6 to 50 minutes in the latter. This difference in the length of the time of the feeding-locomotion may perhaps be due to the difference of species, but on the other hand it may also be due to differing conditions of environment. Namely, *S. atra* usually creeps on rocks exposed to the direct sunlight of the tropics, the surface of these rocks desiccating more rapidly than the submerged rocks in 40° 55' N-L, where *S. japonica* is commonly found. The supposition above mentioned seems to be all the more reasonable when we observe the fact that *S. atra* creeps for a longer period in the night-time than in the daytime.

3. The homing habits.

Concerning the homing habits of molluscs, they have been well studied in the case of *Patella vulgata*, and DAVIS (1895) says "Observations confirmed the statements previously made by various naturalists, from Aristotle onwards, that the common limpet (*Patella vulgata*) settles down on some eligible spot (its "scar") between tide-marks, and makes a home, to which it returns after having been out to feed." And according to RUSSELL (1909), it is said that the following writer have studied the habits of limpets; LUKIS (1831), BOUCHARD-CHANTERAUX (1883), DAVIS (1885, 1894), FISCHER, P. (1887), MORGAN (1894), FISCHER, H. (1898) and WILLCOX (1905). Afterwards, PIÉRON (1909, a, b) studied *Patella* and *Calyptraea*; AREY and CROZIER (1918, 1921) studied *Onchidium* (*Onchi-*

della) *floridanum* DALL; CROZIER (1921) studied *Chiton tuberculatus*; ORTON (1915, 1929) studied *Patella vulgata*; ABE (1931, '33) studied *Acmaea dorsuosa* GOULD, and *Siphonaria atra* QUOY et GAIRD and *S. siphon* SOWERBY (ABE, 1935, a, '39, a), and also other species of *Siphonaria* sp. (ABE, 1935, b). And the writer may add here that *Cellana mauritiana* (PILSBRY) and *Patelloida saccharina* (LINNÉ) inhabiting the lime-stone rocks at Palao (ABE, 1937, b) also mark their scar or home on the rock very clearly.

In the methods of homing, there may exist some gradations as AREY and CROZIER (1921) already indicated by "Beginning with *Chiton tuberculatus* (CROZIER, '21), in which there can be found something like 'homing', but of a rather vague type and pretty certainly the results of immediate stimulation, a series comprising also *Patella*, *Onchidium*, and *Octopus* exhibits more and more highly developed 'homing' propensities. The return of a *Patella*, *Fissurella*, *Siphonaria*, or *Calyptrea* to its specific site cannot be accomplished beyond a relatively slight distance; these creatures also tend to follow fairly definite paths in their excursions and to adhere to these paths when returning; and some of them creep but slightly, if at all, away from their scars. *Onchidium*'s behaviour is obviously an advance in respect to complexity. Analogous behaviour has been described for snails and slugs (as in the famous story of the sick snail and its companion, cited by DARWIN, '71, P. 316, and by other; cf. also COOKE, '95, and SCHARFF, '07). The investigation of this matter in snails and slugs holds the possibility of considerable interest. Finally, the behaviour of *Octopus* (cf., e. g., COWDRY, '11), which returns to its nest after extensive forays and from considerable distances, under circumstances such that direct vision of the nest entrance is completely excluded, represents the most complex form of this activity among mollusc." And they conclude by saying "There is no evidence of associative or persisting memory in connection with homing, nor do other activities of *Onchidium* point to the existence in the form of anything approaching intelligent behavior. Responses to immediate stimulations are adequate for the analysis of the situation." FLATELY and WALTON (1922) said, "Before having recourse to explanations involving higher mental activities it is well to consider the part played by the environment. Thus BOHN (1909) finds that 'homing' is not always certain. The movements of the animal seem to be influenced by gravity. There are lines of least resistance on the rock which are followed very much as one might follow them in a forest. It is not necessary to invoke a muscular or visual memory."

On the other hand, RUSSELL (1934) said emphasizing the opinion of PIÉRON (1909), "It is clear that it (*Patella*) does not regain its home purely by chance or by blind trial and error", and "there is an active and determined effort to get back to and re-occupy the customary niche."

WILLCOX (1905) speaks of the 'homing' of *Siphonaria alternata* and of *Fissurella barbadensis*, both occurring on the calcareous rocks lying between tide-marks at Bermuda. "The former did not 'home' if removed more than six inches, but generally returned if removed more than six inches, but generally returned if moved a couple of inches away, doing best in quiet shallow tide pools. If headed away from the scar *Siphonaria* turns of its own accord. One specimen formed a new home and made a green spot in three days. The range for *Fissurella* is about two inches, but otherwise its behaviour is very much like that of *Siphonaria*."

Siphonaria atra QUOY et GAIMARD (ABE, 1935, a, '39, a) also has a home and returns to it after journeys made to spots about from 6 to 30 cm distant. And on the way home, the limpet creeps along a track close to the previous path so close that it is reach of its mantle margin. If artificially removed from its home, up to a distance of 15 cm, the limpet usually will return, some have even been known to return home even when artificially removed a distance of 30 cm, but it is understood that in these cases the limpets had been able to find food paths marked by themselves in previous journeys. Nearly the same results were obtained also in the case of *Siphonaria siphon* SOWERBY.

In the case of *Siphonaria japonica* DONOVAN, the distance of the journey is limited to within about 15 cm away from the home, thus it is longer than that made by *Siphonaria alternata* and shorter than that by *S. atra* but about equal to that made by *S. siphon*. The habits of the limpets on the way home are quite the same in the cases of *S. atra* and of *S. japonica*. The characteristics of *S. japonica* are that it rests at its destination for certain minutes, and that it has the habit of making secondary and tertiary journeys for more feeding.

Such characteristic behaviour may perhaps be suitable to this kind of limpet inhabiting rocky shores of northern seas where the waves beat more violently than in the habitats of *S. atra* and of *S. siphon*. And it is noticeable that the limpet begins its secondary or tertiary journey only after it has definitely returned to its home after the first journey. This fact seems to suggest the method of homing, viz. the limpet can return home only by retracing the path travelled in the previous journey, or by creeping very close to it.

The limpet's methods of retracing its tracks on the way home have been enquired into by many writers. PIÉRON (1909, a, b) states of *Patella vulgata* that the limpet must have a muscular memory of the direction and distance it has travelled, and that this topographical knowledge is a tactile knowledge acquired mainly by means of the feelers on the head. But the writer cannot accept such a theory of the existence of a memory concerning the direction and distance of the outward journey, from experiments made on *Siphonaria atra* and *S. sipho* (ABE, 1939, a). And it is an important fact that the limpet creeps homewards taking a slightly different path from the outward track, though it is made very close to the latter. PIÉRON then, and those who hold a similar opinion, must be corrected, at least in the cases of *Siphonaria atra*, *S. sipho* and *S. japonica*.

It is conceivable that the method taken in returning home is quite simple in that the limpet creeps very close to the outward track, and the latter may be easily found by using the tentacles on the head and the mantle edge. The reason for taking a different course on the way home is that the limpet will find more food than it would on the original track where it has already been feeding.

4. The periods of mating and spawning.

FLATTELY and WALTON (1922), say "The period of reproductive activity is frequently adjusted to the seasons, or may even coincide with a particular phase of the moon (e.g. *Convoluta*), the result being to allow the larvae the optimum chance of survival."

The breeding season of *Siphonaria japonica* DONOVAN is during the months of May, June and July at Asamushi, and the periods of spawning are clearly related to lunar periodicity. This phenomenon of breeding occurring in relation to lunar periodicity is widely known in the cases of animals and plants, and they are listed in Table IV.

Besides the species listed in Table IV, several algae belonging to the Family Fucaceae exist that produce their sexual cells at about full-moon or new-moon in the neighbourhood of the Asamushi Station (according to Mr. KÔGORO ABE). The Kanaka tribes say that the robber-crab, *Birgus latro* LINNAEUS, spawns its eggs between the peliopods (the peliopods are entirely absent in the male) at the time of the new-moon at Peliliu, South Sea Islands, and Mr. TATSUO AOKI says that certain sand-flies come into the house in groups at the time of the new-moon, while they disappear completely at the time of the full-moon at Peliliu. Though there exist many animals and plants which breed in relation to

TABLE IV. Plants and animals in which breeding is known to occur in relation to lunar periodicity.

Species name	Phase of moon	Localities	Author
Algae			
<i>Neoderma</i>	bilunar	Helgoland	KUCKUCK (1901)
<i>Dictyota dichotoma</i>	bilunar	Plymouth Bangor	WILLIAMS (1905)
<i>Dictyota dichotoma</i>	lunar	Beaufort N. Carolina	HOYT (1907)
<i>Sargassum enerve</i>	bilunar	Misaki	TAHARA (1909)**
<i>Dictyota dichotoma</i>	bilunar	Naples	LEWIS (1910)
<i>Dictyota dichotoma</i>	bilunar	Europe	HOYT (1927)*
"	full moon	N. Carolina	"
<i>Dictyota dentata</i>	full moon	Jamaica	"
<i>Dictyota ciliolata</i>	full moon	Jamaica	"
Coelenterata			
<i>Obelia geniculata</i>	full moon	Millport	ELMHURST (1925)
<i>Pocillopora bulbosa</i>	full and new	Great Barrier Reef	MARSHALL & STEPHENSON (1933)*
<i>Fungia actiniformis</i> var. <i>palawensis</i>	new moon	Palao	ABE (1937, a)*
Echinodermata			
<i>Toxopneustes variagatus</i>	full moon	Tortugas	TENNETT (1910)
<i>Centrechinus (Diadema)</i> <i>setosus</i>	full moon	Suez	FOX (1923)
Polychaeta			
<i>Leodice viridis</i>	third quarter	Samoa	WHITMEE (1875)
<i>Leodice fucata</i>	third quarter	Tortugas	MAYER (1900)
<i>Ceratocephale osuawai</i>	new and full	Tokyo	IZUKA (1903)*
<i>Convoluta roscoffensis</i>	new and full	Brittany	GAMBLE & KEEBLE (1903)
<i>Lysidice oele</i>	full		HORST (1905)
<i>Amphitrite ornata</i>	new and full	Woods Hole	SCOTT (1909)
<i>Nereis dumerilii</i>	1st & 3rd quarter	Naples	HEMPelman (1911)
<i>Odontosyllis enopla</i>	third quarter	Flatt's Is.	GALLOWAY & WELCH (1911)
<i>Nereis limbata</i>	full to 3rd q. 3rd q. to new	Woods Hole	LILLIE & JUST (1913)
<i>Platynereis megalops</i>	between full and new	Woods Hole	JUST (1914)
<i>Eulalia punctifera</i>	third quarter	Conareneau	FAGE & LEGENDRE (1926)
Mollusca			
<i>Chiton tuberculatus</i>	full	Bermuda	CROZIER (1920)
<i>Chaetopleura apiculata</i>	between full and 3rd q.	Woods Hole	GRAVE (1922)

Species name	Phase of moon	Localities	Author
<i>Ostrea edulis</i>	full	Falmouth	ORTON (1926)*
<i>Cumingia tellinoides</i>	full	Woods Hole	GRAVE (1927)*
<i>Pecten opercularis</i>	full	Plymouth	AMIRTHALINGAM (1928)*
<i>Acanthozostera gemmata</i>	full	Great Barrier Reef	STEPHENSON (1934)*
<i>Siphonaria atra</i>	half moon	Palao	ABE (1935, a, '39, a)*
<i>Siphonaria siphon</i>	half moon	Palao	ABE (1935, a, '39, a)
<i>Melarhaphe (Littorinopsis) scabra</i>	half moon	Palao	ABE (1936, '39, b)
<i>Siphonaria japonica</i>	full moon	Asamushi	ABE (1939 a)
Pisces			
<i>Leuresthes tenuis</i>	full and new	California	THOMPSONS (1919) CLARK (1925)
Mammalia			
<i>Homo sapiens</i>			ARRHENIUS (1898)

* The writer made observation in 1909 of the periodicity occurring in the oögonium liberation, and found that the intervals between two successive liberations vary in an irregular manner, without having any fixed relation to the highest spring tide (TAHARA, 1913).

* Papers other than these were cited mainly from AMIRTHALINGAM's paper of 1928.

lunar periodicity, in most cases the causes are not clearly explained.

It is said by IZUKA (1903) that the Japanese Palolo, *Ceratocephale osawai* IZUKA swarms most abundantly within 3 days after the new-moon and after the full-moon in the months of October and November, and that the swarming is greater after the new-moon than after the full-moon, and moreover the same writer noticed that the spring-tide following the new-moon is higher than that which comes after the full-moon, remarking that "There is then a noticeable parallelism between the occurrence of the densest swarm and the highest spring-tide during the months concerned."

ORTON (1926) who studied *Ostrea edulis* listed and described the probable predisposing factors of periodicity in the spawning as follows: "1. Tide, variation in height, accompanied by increase and decrease of pressure, temperature, salinity and other more recondite hydrographical factors, such as pH at successive high and low tides. In spring-tides the rates of change in the above factors will generally be greatest, so that decrease of pressure, increase of temperature and to a less extent decrease of salinity will act together. 2. Moonlight: variation in intensity and duration. Maximum values will occur about new moon spring tides.

3. Food: variation in the amount of (a) available food materials, and/or (b) food-intake in the tidal or bi-lunar cycle. Food may be most abundant and feeding most active during neap-tides, but this is not known with accuracy, but in view of the rapid development of the sex-elements these factors are undoubtedly important. 4. Temperature: absolute variation. There can be little doubt that a temperature of about 60° F constitutes the lower limiting value for breeding in the oyster. 5. Sunshine: the variation in duration and intensity of which should not be neglected in a study of an estuarine form: maximum values on the beds would tend to occur at spring tides. 6. Undetected factors, such as may have operated in the course of evolution, and may or not be determinable."

GRAVE (1927) says with regard to *Cumingia tellinoides*, "The heaviest spawning occurs at the period of the full moon until new moon, and that the period of the first quarter is the period of restricted spawning. This behavior of *Cumingia* can scarcely be explained on any other ground than as a lunar effect." and "It is shown that temperature is not the only factor which determines the duration of the spawning season and periods of spawning."

HOYT (1927) says on an algae, *Dictyota*, that "The fruiting periods seem to be related to the tides, but the relation of fruiting period to tide is different for each region studied. Wherever the tide are regular, as in England, Wales, Italy and North Carolina, whether range is greater or smaller, the periods are regular and constant unless retarded by unfavorable conditions; but where the tides are irregular, as in Jamaica, the periods are scarcely evident; Where the range of tide is very slight, as at Naples, the development of the fruiting crops is less uniform than where the range is greater, as in Wales, England, and North Carolina."

AMIRTHALINGAM (1928) studied *Pecten opercularis*, and says "Hence it would appear that there is a maximum temperature limit of about 11° C for *Pecten opercularis* which the breeding of the species stops. This, with the work of Orton and Fox, suggests that there is a maximum and minimum temperature limit for each species of the marine animals, between which temperatures breeding mainly occurs." and "It is suggested that in the animal there is a physiological rhythm that causes the development of the gonad to coincide with the full moon of each lunar month."

The writer has studied *Siphonaria atra* QUOY et GAIMARD (ABE, 1935, a, '39, a) and *Melarhaphé (Littorinopsis) scabra* (LINNAEUS) (ABE, 1936, '39, b) and has found that the former species mate in the days at the time of the full-moon and perhaps also at the time of the new-moon,

and spawn at the time of the half-moon, but the later species mate mainly at the time of the half-moon, though this species lives on the mangrove-tree growing in the littoral zone. On the other hand, the writer has studied *Batillaria multiformis* (LISCHE) (ABE, 1934), and has found that this species shows a rhythmical change in its movement and migration, and at the time of the half-moon, the number of individuals to be found are most numerous, while at the time of the full-moon and new-moon, they are few in number. Considering the above facts, the writer has taken particular note of locomotion, of mating and of migration, and has stated his opinion of the importance of the combination of the factors of tidal-phase and change of light-intensity by day and by night, and the result of this combination appears to be related to lunar periodicity (ABE, 1936, '39, b).

Now considering *Siphonaria japonica*, the mating of this species takes place or begins to take place mainly at the time of the waxing half-moon, and the spawning takes place or begins to take place mainly at the time of the full-moon. While both *Siphonaria atra* and *S. sipho* mate at the time of the full moon, and perhaps also at the time of the new-moon, and spawn at the time of the half-moon. Furthermore, individuals of the former species are seen mating in the morning or forenoon while in the latter they are seen doing so mainly in the night-time. Therefore it may be considered that *Siphonaria japonica* has the habit of mating when the light is weak, such as in the twilight, and *Siphonaria atra* or *S. sipho* has the habit of mating in the darkness. Therefore the time of mating may be decided according to factors both of light intensity and time of low-tide, as the two *Siphonarias* creep in the open air only at low-tide.

At Palao, where *Siphonaria atra* and *S. sipho* are found, the low-tide occurs at mid-night at the times of the full-moon and of the new-moon, and consequently the period of the mating of these species may be definitely said to occur mainly at the time of the full-moon or of the new-moon. At Asamushi, the habitat of *Siphonaria japonica*, low-tide occurs at daybreak or at sun-set a few days after the half-moon, and thus the period of the mating of this species may be said to occur mainly on the days following the half-moon.

Here it must be noticed that there is a maximum and minimum limit of temperature for each species living in the temperate regions, such as in the case of *Siphonaria japonica*. But the conception of the combination of the factors of tide-phase with the intensity of light by day and

by night may explain throughout to some extent the phenomena which occur in connection with lunar periodicity.

5. The egg and its development.

Here I should like to compare the egg and larva of *Siphonaria japonica* in various developmental stages with those of other species of *Siphonaria*.

The egg-ribbon of *Siphonaria japonica* is nearly semicircular or nearly circular in form enveloped in a thick gelatinous mass, and it resembles those of *S. lepida* GOULD (FUJITA, 1904, Pl. 1, Fig. 1), *S. australis* QUOY and GAIMARD (HUTTON, 1882, Pl. 15, Fig. 8) and of *S. siphon* SOWERBY. In size, it is nearly the same as those of *S. japonica*, *S. siphon* and *S. lepida*. The egg-ribbon of *Siphonaria atra* (ABE, 1939, a, Fig. 9) is an irregular whirlpool in shape enclosed in a tape-like gelatinous mass, and the longer diameter attains to about 2.8–3.4 cm. Thus the form of the egg-ribbon of *S. atra* is quite different from those of the other species of *Siphonaria*.

The eggs of the *Siphonaria* are enveloped in an egg-capsule of elipsoidal form, and are linked one to another by means of fine threads attached to both ends. The size of the egg-capsule is about 0.18 mm in the longer diameter (L) and is 0.1 mm in the shorter diameter (B) in *S. australis*; 0.18 mm (L), 0.13 mm (B) in *S. atra*; 0.27 mm (L), 0.17 mm (B) in *S. lepida* and 0.27 mm (L), 0.15 mm (B) in *S. japonica*. The diameter of the egg is about 0.067 mm in *S. australis*, 0.085 mm, in *S. atra*, 0.10 mm, in *S. lepida* and 0.087 mm to 0.091 mm in *S. japonica*.

The shape of the post-larva, seen just before hatching, is nearly the same in both species of *S. atra* and *S. japonica*, but the size of the larval shell is about $146\mu \times 98\mu$ in *S. japonica* and about $84\mu \times 55\mu$ in *S. atra*. Therefore the larva just before hatching is larger in the *Siphonaria* living in the temperate zones than in the *Siphonaria* living in the tropical seas, though the size of the shell of the adult individual is smaller in the former species.

Of the development of the larva, in *S. atra* it hatches out about 4 or 5 days after spawning in the tropical seas (Temperature of seawater is about 27–28°C) while in *S. japonica* it hatches out about 15–20 days after spawning in the temperate zone (Temperature of seawater is about 13–15°C) and this difference in the period of hatching is probably caused by the difference in the temperature of the seawater. On the other hand, it must be noticed that the spawning is seen about 7 days after the mating in both *S. atra* and *S. japonica*.

As is suggested in the above, it would be an interesting and important problem to study the differences in the periods between the mating and the development of the larva of animals living in tropical and in colder seas.

SUMMARY

1. *Siphonaria japonica* DONOVAN inhabits the level just below the mean-tide-level in the warmer season, though its zonation is different according to the influence of wave action, and it migrates a little below this level in the winter time. The relationship existing between the zonations of *Siphonaria japonica* and other animals and plants inhabiting the littoral zone is studied in the present paper.

2. The *Siphonaria* begins to creep for feeding purposes before or just after the time when it is exposed to the air during the ebbing-tide, and continues locomotion for about two hours, and it does not usually creep in the water. It is found that the phenomenon exists of a sequence of the times of creeping in the case of animals living in limited areas in the littoral zone.

3. The food of the *Siphonaria* consists mainly of algae but it also eats the sedentary Diatoms. The latter does not seem to serve as food as they are excreted by this animal while they are still living.

4. The *Siphonaria* forms a distinct home, and shows a homing instinct. The distance of locomotion for feeding is limited in general to within about 15 cm. And this species makes sometimes a second or even a third journey for feeding after it has definitely returned home from the first journey. The individuals of a smaller size with a shell shorter than about 6 mm sometimes do not show any homing instinct.

5. The method of the return home in the case of the *Siphonaria* is to creep very close to the outward tracks, which are easily followed by means of tentacles on the head and by using the mantle margin.

6. The *Siphonaria* is a hermaphrodite animal, mating mainly at the time of the waxing-moon. In the act of mating, the heads of two individuals come into contact and the penis is extruded emitting the spermatophore.

7. The spawning takes place about 7 days after the time of mating and therefore it occurs at the time of the full-moon. The species spawn the egg-ribbon on rock-faces on a level situated just below its habitat.

8. The eggs are enveloped in egg-capsules of ellipsoidal form, and are linked one to another by a fine filament attached to both ends of

the longer axis. The larva is retained within the egg-capsule till it develops to the stage of post-larva, and then it hatches out about 15 to 20 days after the spawning, and therefore it occurs at the time of the new-moon.

9. The spawning season takes place during the months of May, June and July at Asamushi, where the temperature of the seawater ranges from 13°C to 23°C.

10. The phenomenon of periodicity, which is noticed in the breeding and other habits, and which occurs in relation to lunar periodicity, seems to be explained by the conception of the combination of the factor of the tidal-phase with the factor of the intensity of light, either of day in which the animals creep most actively, or of night in which they do not creep at all.

REFERENCES

- ABE, NORORU (1931). Ecological Observations on *Acmaea dorsuosa* GOULD. Sci. Rep. Tôhoku Imp. Univ., 4th Ser., Biol., Vol. VI, No. 3, 1931, pp. 403-427.
- ABE, N. (1932). The Age and Growth of the Limpet (*Acmaea dorsuosa* GOULD). Ibid. Vol. VII, No. 3, 1932, pp. 347-363.
- ABE, N. (1933). The Colony of the Limpet (*Acmaea dorsuosa* GOULD). Ibid. Vol. VIII, No. 2, 1933, pp. 169-187.
- ABE, N. (1934). Ecological Observations on *Batillaria multiformis* (LISCHE). Ibid. Vol. VIII, No. 4, 1934, pp. 383-398.
- ABE, N. (1935, a). On the Locomotion of a Limpet-like Pulmonata, *Siphonaria atra* QUEY et GAIMARD. The Venus, Vol. V, No. 4, 1935, pp. 206-213. (in Japanese with Summary in English)
- ABE, N. (1935, b). On the Direction of the Attachment of Limpet (*Siphonaria*) and of Sedentary Organisms (*Navicula* and *Tetracultis*). Ecol. Rev., Vol. 1, No. 3, 1935, pp. 207-211. (in Japanese)
- ABE, N. (1935, c). The Colony of the Littorina: *Littorivaga brevicula* (PHILIPPI). Sci. Rep. Tôhoku Imp. Univ., 4th Ser., Biol., Vol. IX, No. 4, 1935, pp. 279-296.
- ABE, N. (1936). Ecological Observations on *Melurhaphe (Littorinopsis) scabra* LINNAEUS, mainly on its Locomotion. Ecol. Rev. Vol. 2, No. 1, pp. 35-42, No. 2, pp. 132-136. (in Japanese)
- ABE, N. (1937, a). Post-larval Development of the Coral *Fungia actiniformis* var. *palauensis* DÖDERLEIN. Palao Trop. Biol. Sta., Studies No. 1, 1937, pp. 73-93.
- ABE, N. (1937, b). Ecological Survey of Iwayama Bay, Palao. Ibid. No. 2, 1937, pp. 217-324.
- ABE, N. (1938). Types of Locomotion of Gastropods and the Relation to the Time of Locomotion and to the Habitats. Ecol. Rev. Vol. 4, 1938, pp. 51-60. (in Japanese with Summary in English)
- ABE, N. (1939, a). Ecological Observations on a Limpet, *Siphonaria atra* QUEY et GAIMARD, with a Note on Homing Behaviour of Limpets. Palao Trop. Biol. Sta., Studies No. 6. (in Press)
- ABE, N. (1939, b). Ecological Observations on *Melurhaphe (Littorinopsis) scabra* LINNAEUS

Inhabiting on the Mangrove-tree. *Ibid.* No. 6. (in Press)

- AMERTHALINGAM, C. (1928). On Lunar Periodicity in Reproduction of *Pecten opercularis* near Plymouth in 1927-28. *Journ. Mar. Biol. Assoc. (N. S.)* Vol. 15, 1928, pp. 605-641.
- AREY, L. B. and CROZIER, W. J. (1918). The 'Homing Habits' of the Pulmonate Mollusk *Onchidium*. *Proc. Nat. Acad. Sci.*, Vol. 4, 1918, pp. 319-321.
- AREY, L. B. and CROZIER, W. J. (1919). The Sensory Responses of Chiton. *Journ. Exp. Zool.*, Vol. 29, 1919, pp. 157-260.
- AREY, L. B. and CROZIER, W. J. (1921). On the Natural History of *Onchidium*. *Ibid.* Vol. 32, No. 3, 1921, pp. 443-502.
- BATCHELDER, J. (1915). Migration of *Ilyanassa obsoleta*, *Litorina litorea* and *Lit. rudis*. *Nautilus*, Vol. 29, 1915. (cited from LINKE, 1935)
- BOHN, G. (1905). Attractiveness et Oscillations des Animaux Marin sur l'Influence de la Lumiere. *Inst. Generale Psychologique*, Paris. (cited from HASEMAN, 1911)
- BOHN, G. (1909). De l'Orientation chez les Patelles. *C. R. Acad. Sci.*, T. 148, pp. 868-870. (cited from FLATTERLY and WALTON, 1922 and AREY and CROZIER, 1921)
- BOKENAM, N. A. H., NEUGEBAUER, F. L. M., and STEPHENSON, T. A. (1938). The Vertical Distribution of Certain Intertidal Marine Gastropods in False Bay, with Notes on the Development of Two of them. *Ann. Natal Mus.*, Vol. 9, pp. 113-138.
- BRIGHT, K. M. F. (1938). The South African Intertidal Zone and its Relation to Ocean Currents. II. An Area on the Southern Part of the West Coast. *Transact. Royal Soc. South Africa*, Vol. 26, 1938, pp. 49-66.
- CROZIER, W. J. (1921). "Homing" Behavior in Chiton. *Amer. Naturalist*, Vol. 55, 1921, pp. 276-281.
- DAVIS, R. J. (1885). The Habits of the Limpet. *Nature*, Vol. 31, 1885, pp. 200-201.
- DAVIS, R. J. A. (1895). The Habits of Limpets. *Ibid.* Vol. 51, 1894-95, pp. 511-512.
- EYRE, JOYCE (1939). The South African Intertidal Zone and its Relation to Ocean Currents. VII. An Area in False Bay. *Ann. Natal Mus.*, Vol. 9, pp. 283-306.
- FLATTERLY, F. W. and WALTON, C. L. (1922). The Biology of the Seashore. London, 1922, p. 336.
- FUJITA, T. (1895). Preliminary Note on the Mesoderm Formation of Pulmonata. *Zool. Magaz.* Vol. 7, 1895, pp. 89-93.
- FUJITA, T. (1904). On the Formation of the Germinal Layers in Gastropoda. *Journ. Coll. of Sci. Imp. Univ. Tokyo*, Vol. 20, Art. 1, 1904, pp. 1-42.
- GRAVE, BENJAMIN, H. (1927). An Analysis of the Spawning Habits and Spawning Stimuli of *Cumingia tellinoides*. *Biol. Bull.*, Vol. 52, 1927, pp. 418-435.
- HASEMAN, J. D. (1911). The Rhythmical Movements of *Litorina litorea* Synchronous with Ocean Tides. *Biol. Bull.*, Vol. 21, 1911, pp. 113-121.
- HEATH, H. (1899). The Development of *Ischnochiton*. *Zool. Jahrb. Abt. Anat. Bd.* 12, pp. 1-90. (cited from CROZIER, 1921).
- HOYT, W. D. (1927). The Periodic Fruiting of *Dictyota* and its Relation to the Environment. *Amer. Journ. of Botany*, Vol. 10, 1927, pp. 592-619.
- HUTTON, F. W. (1882). Notes on the Structure and Development of *Siphonaria australis* QUEY and GAIMARD. *Annals and Magazine of Natural History*, Ser. 5, Vol. 9, No. 53, 1882, pp. 341-344.
- LEUKA, AKIRA (1903). Observations on the Japanese Palolo, *Ceratocephale osawai*, n. sp. *Journ. Coll. Sci. Imp. Univ. of Tokyo Japan*, Vol. 17, Art. 11, 1903, pp. 1-37.
- LINKE, OTTO (1935). Der Leich von *Litorina (Melaraphe) neritoides* L. *Zoologischer An-*

- zeiger, Bd. 112, 1935, pp. 57-62.
- LINKE, OTTO (1939). Die Biota des Jadebusenwattes. Helgoländer wissenschaftliche Meeresuntersuchungen. Band 1, Heft 3, pp. 201-348.
- MARSHALL, S. M. and STEPHENSON, T. A. (1933). The Breeding of Reef Animals, Part 1. The Corals. Great Barrier Reef Expedition 1928-29, Sci. Rep., Vol. 3, No. 8, 1933, p. 219-245.
- MITSUKURI, KAKICHI (1901). Negative Phototaxis and Other Properties of *Littorina* as Factors in Determining its Habitat. Ann. Zool. Japonenses, Vol. 4, Part 1, 1901, pp. 1-19.
- MORGAN, C. L. (1894). The Homing of Limpets. Nature Vol. 51, 1894-95, p. 127.
- MORI, SYUITI (1937). Studies on the Annual Vertical Migration and the Life History of a Japanese Fresh-water Snail, *Melanoides (Semisulcospira) niponicus* (SMITH). Zool. Magaz., Vol. 49, No. 2, 1937, pp. 37-50.
- MORSE, MAX WITHROW (1910). Alleged Rhythm in Phototaxis Synchronous with Ocean Tides. Proc. Soc. for Exp. Biol. & Med., Vol. 7, 1909-10, p. 145.
- ORTON, J. H. (1915). On the Breeding Habits of *Echinus miliaris*, with a Note on the Feeding Habits of *Patella vulgata*. Journ. Mar. Biol. Assoc., Vol. 10 (N. S.), 1913-15, pp. 256-257.
- ORTON, J. H. (1926). On Lunar Periodicity in Spawning of Normally Grown Falmouth Oysters (*O. edulis*) in 1925, with a Comparison of the Spawning Capacity of Normally Grown and Dumpy Oysters. Ibid. Vol. 11 (N. S.), 1926-27, pp. 199-225.
- ORTON, J. H. (1929). Observation on *Patella vulgata*. Part III. Habit and Habitats. Ibid. Vol. 16 (N. S.), 1929-30, pp. 277-288.
- PEARSE, A. S. (1936). The Migrations of Animals from Sea to Land. Durham, 1936, p. 336.
- PIÉRON, H. (1909, a). Contribution à la Biologie de la Patelle et de la Calyptrée, Le Sens du Retour et la Mémoire Topographique. Archives Zool. Exp. et Générale, 5^{me} Ser., T. 1, 1909, pp. 18-29.
- PIÉRON, H. (1909, b). Contribution à la Biologie de la Patelle et de la Calyptrée. Bull. Sci. Fr. et Belg., T. 43, 1909, pp. 183-202. (cited from AREY and CROZIER, 1921, and RUSSELL, 1934)
- RUSSELL, E. S. (1909). Environmental Studies on the Limpet. Proc. Zool. Soc. of London, Vol. 2, 1909, pp. 856-870.
- RUSSELL, E. S. (1934). The Behaviour of Animals, an Introduction to its Study. London, 1934, p. 184.
- STEPHENSON, ANNE (1934). The Breeding of Reef Animals. Part II. Invertebrates other than Corals. Great Barrier Reef Expedition 1928-29, Sci. Rep., Vol. 3, No. 9, 1934, pp. 247-272.
- STEPHENSON, T. A. (1936). The Marine Ecology of the South African Coasts, with Special Reference to the Habits of Limpets. Proc. Linn. Soc. Lond., Vol. CXLVII, pp. 74-79.
- STEPHENSON, T. A. (1939). The Constitution of the Intertidal Fauna and Flora of South Africa. Part I. Journ. Linn. Soc. Zool., Vol. XL, pp. 487-536.
- SUZUKI (TANITA), SENJI (1935). Notes on the Behavior of *Nerita japonica* DUNKER. Sci. Rep. Tôhoku Imp. Univ., Fourth Ser., Biology, Vol. 9, No. 4, 1935, pp. 297-314.
- TAHARA, MASATO. (1909). On the Periodical Liberation of the Oospheres in *Sargassum*. Bot. Mag., Tokyo., Vol. 23, 1909, p. 151.

- TAHARA, MASATO. (1913). Oogonium Liberation and the Embryogeny of Some Fucaceous Algae. Journ. Coll. Tokyo Imp. Univ., Vol. 32, Art. 9, 1913, pp. 1-13.
- WILCOX, M. A. (1905). Homing of *Fissurella* and *Siphonaria*. Science, N. S. Vol. 22, 1905, pp. 90-91. (cited from PIÉRON, 1909, and FLATTERLY and WALTON, 1922.)

DIURNAL VARIATION IN THE BODY TEMPERATURE OF
THE STRAWBERRY WEEVIL, *ANTHONOMUS*
BISIGNIFER SCHENKLING

(THE DIURNAL RHYTHM OF ACTIVITIES IN INSECTS AND
ITS ENVIRONMENTAL CONDITIONS NO. VIII)

By

MUTSUO KATÔ

Biological Institute, Tôhoku Imperial University, Sendai

(With 4 text-figures)

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INTRODUCTION

The writer published previously several papers concerning the diurnal activity of insects relating to its environmental conditions. In the present paper the diurnal variation in the body temperature of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING, is dealt with.

From the writer's previous papers (KATÔ, 1936, 1937 a, b, 1938 a, b) concerning the ecological investigation on the diurnal activity of the Strawberry Weevil, it was recognized that some close correlation exists between the activities of the Strawberry Weevil and the environmental temperature factors, and that the diurnal rhythm occurring in the activity of the said weevil is mainly governed by various temperature factors, and thus was proposed the necessity of directing our attention towards the ecological investigation of the body temperature of this weevil.

The writer had measured the body temperature of the same weevil in field condition (KATÔ, 1939). Considering the results obtained from this investigation, it was concluded that the process of the fluctuation of the body temperature is caused primarily by the increase and the decrease of the solar radiant energy and is affected only secondarily by other meteorological factors. Of course, it will be needless to say that the body temperature is based upon the air temperature and, therefore, the solar radiant energy is effective in an additional condition to the air temperature.

But we are still not convinced of some close correlation existing between the activity of the Strawberry Weevil and the environmental con-

ditions, because the diurnal variation of the body temperature has not been experimentally clarified.

In the present investigation the writer wishes to deal with the correlation between the diurnal variation of the body temperature and that of the environmental temperature factors.

Before proceeding further the writer wishes to express his sincere thanks to Prof. Dr. SANJI HÔZAWA for his kind guidance and encouragement given to him and to Assist. Prof. Dr. ISAO MOTOMURA for his valuable suggestions. The writer is also grateful to Prof. Dr. SAEMON-TARO NAKAMURA, the director of the Mukaiyama Observatory, and to Assist. Prof. Dr. YOSIO KATÔ of the same Observatory for their kind assistance given him in using the instruments.

MATERIAL AND METHOD

Some overwintered Strawberry Weevils, that were found active in the strawberry garden of GASEN-EN, situated at Mt. Dainenji, Sendai, and that were collected there, were used as material in the present investigation.

The greater part of the present experiments were carried out at the Biological Institute of the Tôhoku Imperial University and partly at the Geophysical Institute of the same.

The thermo-electric method was adopted to measure the body temperature and the thermopile specially designed was used to learn the difference between the body temperature and the air temperature. The solar radiant energy was measured by means of a solarimeter and a black heliothermometer. The body temperature and the solar radiant energy were recorded respectively using millivoltmeter. The air temperature was observed by ASMAN's respiratory thermometer.

RESULT AND DISCUSSION

1. BODY TEMPERATURE IN THE MORNING: (Text-fig. 1. . . . *Experiment No. 7*).

Before the sunrise the body temperature of the Strawberry Weevil is nearly equal to the air temperature. After the sunrise the body temperature rises rapidly in accordance with the increase of the solar radiant heat, and its rising velocity is fairly great compared with that of the air temperature. Thus the body temperature soon becomes 4°C or more higher than the air temperature. It may be seen in this experiment

that the solar radiant heat is fully utilized. The temperature regulation above mentioned is never seen in the midday or in the evening and thus it may be thought that it depends upon the low air temperature which forms the source of the body temperature.

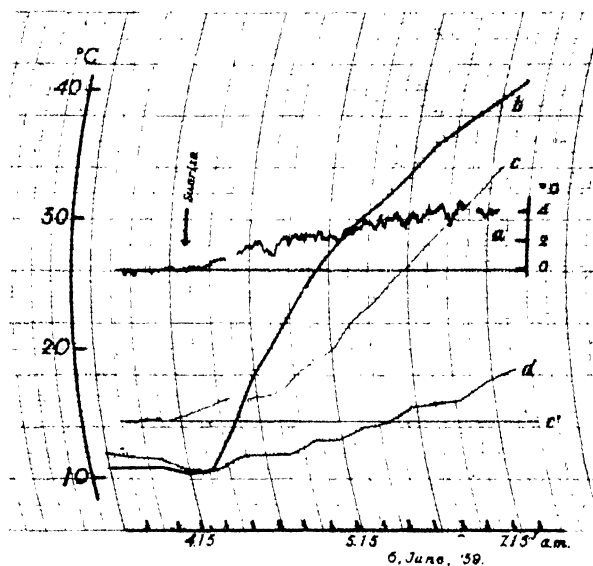


Fig. 1. Diurnal variation of the body temperature of the weevil and the temperature environments. No. 1. a: difference between the body temperature and the environmental air temperature, b: reading of the black heliothermometer, c: solar radiation recorded using a solarimeter, c': zero-line of the solar radiation, d: air temperature (Experiment No. 7.).

The remarkable absorption of the solar energy is seen in the morning due to the low air temperature, and, therefore, if the sun is suddenly obscured by clouds, then the body temperature falls down rapidly to a degree similar to the air temperature showing a phenomenal descent. So it may be permissible to think that the fluctuation of the solar radiation exerts, in the morning, a remarkable influence upon the descent and ascent of the body temperature and therefore that the large difference is seen between the body temperature in the case of the fine morning and that in the case of the cloudy morning, and also that the rising velocity of the body temperature in the former case may be greater than that in the latter case.

II. BODY TEMPERATURE IN THE MIDDAY: (Text-fig. 2.... *Experiment No. 11*).

It seems to be noticeable that the difference between the body temperature and the air temperature is in this period not so large as in the morning showing only 3°C or nearly so. Though the solar energy is the greatest in this time and the air temperature is also the highest and thus it is naturally expected that the body temperature may be fairly higher than the air temperature, the difference between the body temperature and the air temperature was unexpectedly smaller than in the morning as mentioned in the above. Namely the remarkable utilization of the solar radiant heat is not, of course, observed, but the reverse temperature

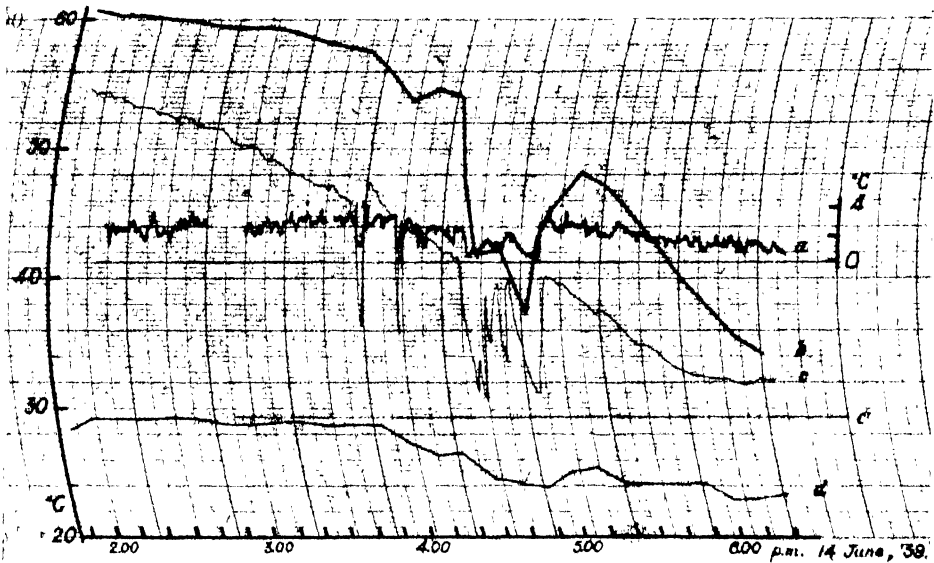


Fig. 2. Diurnal variation of the body temperature of the weevil and the temperature environments. No. 2. (*Experiment No. 11*).

regulation, that is the inhibition of the absorption of the solar radiant heat, seems to be done. This may be evidently caused by the high air temperature forming the base of the body temperature, and, consequently, even if the sunshine is intercepted suddenly by cloud, the body temperature does not show any great fall.

Thus with the rising of the air temperature, the air temperature becomes gradually important as a controlling factor of the body temperature, though in the morning the solar radiant energy was the superior as factor controlling the body temperature.

III. BODY TEMPERATURE IN THE EVENING: (Text-fig. 3 & 4.... *Experiment No. 6 & 9).*

The remarkable utilization of the solar radiant heat which was noticeable in the morning was never observed in the evening. Of course it is needless to say that the temperature regulation due to the high air temperature is not also seen. The difference between the air temperature and the body temperature in this period decreases gradually accompanying with the decrease of the solar radiant energy, and after sunset the body generally keeps a degree of temperature similar to the air.

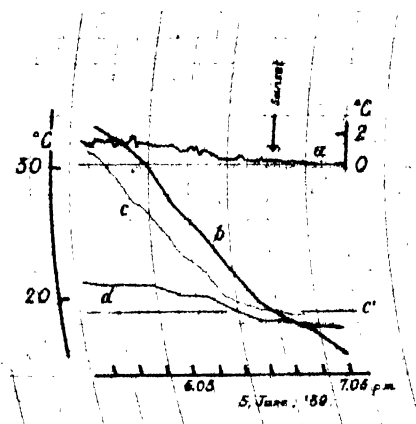


Fig. 3. Diurnal variation of the body temperature of the weevil and the temperature environments. No. 3. (*Experiment No. 6*).

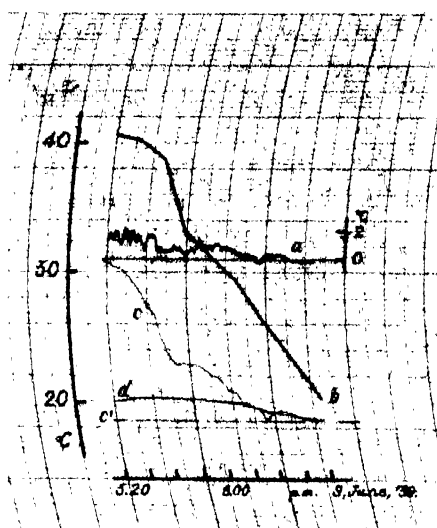


Fig. 4. Diurnal variation of the body temperature of the weevil and the temperature environments. No. 4. (*Experiment No. 9*).

It seems that the phenomenon above mentioned is caused by the fairly high air temperature which forms the base of the body temperature and which is in the optimum temperature zone of the activity of the said weevil. Consequently, even if the sun is obscured by clouds, the body temperature does not fall in a marked degree and is kept still fairly high. And then the body temperature falls gradually being influenced by the falling of the air temperature. It is therefore noticed that the air temperature is one of the most important factors controlling the body temperature.

CONCLUSION

From the results obtained by the experiments above mentioned, we have learned the process of the diurnal variation in the body temperature of the Strawberry Weevil.

The body temperature is generally equal to the air temperature before the sunrise, but after that it rises more rapidly than the air temperature does and becomes 4°C or more higher than the latter. Thus the great utilization of the solar radiant heat is seen.

But with the progression of time this phenomenon becomes weak. In spite of that the solar radiant energy increases rapidly and the air temperature also rises, the absorption of the solar energy to be executed by the body of the weevil decreases. And in the period when the solar radiation is the greatest and the air temperature is also the highest, it is noticed some phenomenon of reverse temperature regulation, that is the inhibition of the absorption of the solar energy, is seen. Consequently the difference between the body temperature and the air temperature becomes only 3°C or nearly so.

In the evening the solar radiation decreases gradually and its amount becomes nearly equal to that in the morning, and thus the evening and the morning stand symmetrically at the noon time in relation to time. Nevertheless, the notable utilization of the solar radiation, which was seen in the morning, is not observed. Of course, the regulation due to the high air temperature, which was observed in the midday, is never seen. This seems to depend upon the fairly high air temperature lying in the optimum temperature zone of activity of the weevil. Body temperature falls gradually accompanying with the gradual decreasing of the solar energy.

In conclusion, regarding the environmental temperature factors which controll the body temperature of the weevil, it may be said that the solar radiant energy is predominant in the morning, but with the progression of time the air temperature becomes important.

The relation between the diurnal variation of the body temperature and the environmental temperature factors has been mentioned above. The body temperature itself is, however, highest in the midday; and then in spite of the remarkable utilization of the solar radiant energy, it is lower in the morning than in the evening, as the air temperature, which forms the base of the body temperature, is fairly lower in the former than in the latter.

It was also known that the rising velocity of body temperature in the morning is very rapidly influenced by the solar energy, but the falling of the same in the evening is rather slow depending upon the gradual fall of the air temperature.

LITERATURE CITED

- KATÔ, M. (1936). On the Activity of the Oviposition of the Strawberry Weevil, *Anthonomus bisignatus* ROELOFS. (The Diurnal Rhythm of Activities in Insects and its Environmental Conditions No. 1). Sci. Rep. Tôhoku Imp. Univ., Biol. Vol. X, 697-708.
- (1937 a). A Statistical Investigation of the Correlation between Climatic Conditions and the Egg-Laying Activity of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING (No. 2). *ibid.*, Vol. XI, 307-321.
- (1937 b). Changes Occurring in the Egg-Laying Activity of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING, in the Case of a Solar Eclipse. (No. 3). *ibid.*, Vol. XI, 353-359.
- (1938 a). The Temperature Limit of Activity of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING. (No. 4). *ibid.*, Vol. XII, 501-510.
- (1938 b). The Diurnal Activity of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING, with a Note of the Ecological Meaning of the Solar Radiant Energy. (No. 5). *ibid.*, Vol. XII, 511-530.
- (1939). Body Temperature of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING, and Its Limiting Factors. (No. 6). *ibid.*, Vol. XIV, 11-19.

ON THE VITAMIN-C (ASCORBIC ACID) CONTENT OF HERBACEOUS PLANTS AND MARINE ALGAE, CONSIDERING FACTORS INFLUENCING IT.

By

TATSUO KOIZUMI AND TOMIO KAKUKAWA

Biological Institute, Tōhoku Imperial University, Sendai

(With one text-figure)

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Since the publication of the easy direct titration method of vitamin-C (TILLMANS *et al.* '30, '32), a large number of papers dealing with vitamin-C has been written by various authors. According to them, vitamin-C occurs or accumulates in large quantities in leaves, flowers, fruits etc. (BESSEY and KING '33, FUJITA and EBIHARA '37, '39 *a*, MITUDA '38 *a*, BESSEY '38 *b*, see also KING '39).

Besides the vitamin-C content of plants used for food, that of marine algae (NORRIS, SIMEON and WILLIAMS '37 *etc.*), wild herbaceous plants (MIWA '38, '39, SAITO and WATANABE '39), leaves of trees (MIWA '38, '39) etc. has been also estimated.

The vitamin-C content of a species is considered to be correlated with various factors, viz., the variety (TRESSLER, MACK and KING '36) the polyploidy (SANSOME and ZILVA '36 *etc.*), the individual, the type of soil (TRESSLER, MACK and KING '36), the fertilizer (ASIKAGA '38, SUGAWARA '38, IJIDO '36), the organic food (REID '38), the season (TRESSLER, MACK and KING '36), the age or maturity (TRESSLER, MACK and KING '36, MITUDA '38 *a*, ASIKAGA '38 *etc.*), the content of chlorophyll (BESSEY and KING '33, GIROUD, RATSIMAMANGA and LEBLOND '34, RANDOIN, GIROUD and LEBLOND '35, ASIKAGA '38) and the light conditions (REID '38, SUGAWARA '39, ASIKAGA '38, MITUDA '38 *b* *etc.*).

As ascorbic acid occurs in relatively large quantities in all green leaves, and both ascorbic acid and chloroplasts reduce AgNO_3 in acid medium, it is concluded that ascorbic acid is associated with chloroplasts (GIROUD, LEBLOND and RATSIMAMANGA '34, GIROUD, RATSIMAMANGA and LEBLOND '34, RANDOIN, GIROUD and LEBLOND '35, GIROUD '38, DISCHEN-

DORFER '37, ASIKAGA '38, WEIER '38, PEKAREK '38 *etc.*), though MIRIMANOFF ('38, '39 *a, b*) questions the correlation between ascorbic acid content and chromoplast pigments (see also CARUSO '38). BUKATSCH ('39) has recently proved the active participation of ascorbic acid in photosynthesis, at least in formaldehyde formation, acting as oxidation-reduction-factors correlated with the function of fluorescent chlorophyll, both in the green plant and in the model (emulsion) in favour of BAUR's scheme ('32, '35, '37 *a, b, c*) while GIROUD ('38) thought that ascorbic acid plays an important part in the condensation process of formaldehyde into sugar.

The present investigation is concerned with (1) the estimation of both reduced and reversibly oxidized forms of ascorbic acid contained in the leaves of wild herbaceous plants, and in marine algae, (2) the effect of light upon the accumulation of ascorbic acid and (3) the relation between ascorbic acid content and variety.

MATERIALS AND METHODS

Materials — Most of the materials used in the present investigation were collected from the Botanical Garden of the Institute. The leaves of the middle parts of the plant were carefully selected, and immediately used for purposes of extraction, those showing defects being discarded.

Extraction and Analysis — As dehydroascorbic acid possesses nearly the same antiscorbutic potency (HIRST and ZUVA '33, FOX and LEVY '36, BORSOOK, DAVENPORT, JEFFEREYS and WARNER '37 *etc.*) and similar participation in photosynthesis as ascorbic acid (BUKATSCH '39), it is necessary to estimate both forms of ascorbic acid.

So-called bound ascorbic acid which may exist in certain plant-tissues and which is not extracted by the usual acid extraction (McHENRY and GRAHAM '35, REEDMAN and McHENRY '38 *etc.*) is considered not to exist (FUJITA and EBIHARA '39 *b*, KATAI '39 *etc.*).

The points to be attended to in the quantitative determination of vitamin-C are considered to be as follows (BESSEY '38 *a, b*, ITO '38, OKUDA and KATAI '39 *etc.*):—

1. The extraction should be complete, and ascorbic acid should be protected from rapid oxidation due to O_2 and enzymes liberated by the macerated cells, or by the presence of traces of certain metals and its reversibly oxidized product, dehydroascorbic acid, from further destruction.—Cold metaphosphoric acid of suitable concentration was used.
2. The indophenol titration should be protected from any interfering substances having a reducing potential lower than the dye.—Titration was carried out near at $0^\circ C$.
3. The reduction of dehydroascorbic acid by H_2S should be complete.—It was carried out in a suitable pH range.
4. Reagents should be protected from rapid change. They were stocked in a refrigerator.

Analytical Procedure.—This was carried out by a method similar to that modified by OKUDA and KATAI (1939). Immediately after being

collected the leaves were washed with distilled water, wiped with filter paper and 2-5 gm. were weighed out. They were placed in a cooled mortar, covered by 5-10 c.c. of cold 10% HPO_3 and then ground up with purified quartz sand within 5 minutes, and diluted with cold water to a volume corresponding to five times the volume of 10% HPO_3 used, or 1:5 dilution (2% HPO_3). After standing in a refrigerator for 30 minutes during which time it was stirred occasionally, the content was centrifuged.

One part of the supernatant fluid (extracts) was used for the determination of (reduced) ascorbic acid; that is, 0.1 c.c. of 0.01 M. 2,6-dichlorophenolindophenol solution, cooled near to 0°C , were titrated with the cold extract from a 2 c.c. messpipette. The standardization of the dye was made by 0.01 N thiosulphate titration of the iodine liberated from KI by the oxidized form of the dye in acid solution (MENAHER and GUERRANT '38). According to BESSY ('38 *b*) sulphuric acid reacts with the dye and should never be used.

The other part of the supernatant fluid was used for total ascorbic acid (ascorbic acid + dehydroascorbic acid): The fluid was treated with H_2S at pH 4.8 (0.6 c.c. 50% Na-acetate to 10 c.c. fluid), and stoppered. After standing overnight, the H_2S was removed by CO_2 or N_2 (controlled by lead acetate paper for several minutes): the solution was made up to twice the volume of the extract by the addition of 10% cold HPO_3 , centrifuged, stoppered, and cooled and the titration was then carried out similarly. Dehydroascorbic acid is represented by the difference between the above two determinations.

RESULTS AND DISCUSSION

1. *Ascorbic acid content of leaves of herbaceous plants.*

As shown in Table I and Fig. 1 the greater part of vitamin-C of the leaves of most herbaceous plants is found in the reduced form. This is the case with animals (KING '39, p. 403, FUJITA and EBIHARA '37, '39 *a* etc.) and other plant leaves (BESSEY '38 *a*, OKUDA and KATAI '38 *etc.*).

TABLE I.
Ascorbic acid (vitamin-C) content of leaves of herbaceous plants.

Material	Family	Date collect. and exp.	Vitamin-C content		Reduced form	Total	per cent
			mg. per gm.	mg. per gm.			
<i>Pennisetum purpurascens</i> MAKINO	Gramineae	1939, Sept. 27	0.754	0.283	1.037	72.7	
<i>Syntherisma sanguinalis</i> DULAC var. <i>ciliaris</i> HONDA		" 28	0.157	0.472	0.629	24.9	
<i>Chaetochloa viridis</i> SCRIBN. var. <i>genuina</i> HONDA		" 29	1.240	0.284	1.524	81.3	
<i>Miscanthus sinensis</i> ANDERS.	Compositae	" "	0.606	0.336	0.942	64.3	
<i>Eupatorium stoeadosum</i> HANCE		" 30	0.695	0.392	1.087	65.9	
<i>Siegesbeckia pubescens</i> MAKINO		Oct. 3	0.370	0.867	1.237	29.9	
<i>Artemisia vulgaris</i> L. var. <i>indica</i> MAXIM.	"	" 6	0.357	0.424	0.781	49.7	
" " (potted)	"	" "	0.309	0.571	0.880	35.1	
<i>Taraxacum henckense</i> NAKAI	"	" 8	0.536	0.230	0.766	69.9	
* <i>Cacalia bulbifera</i> MAXIM.	"	" 6	—	—	0.446	—	
* <i>Cacalia krameri</i> MATSUM.	"	" 14	0.186	0.576	0.762	24.4	
* <i>Cacalia delphinifolia</i> SIEB. et ZUCC.	"	" 28	0.121	0.272	0.393	30.7	
<i>Lespedeza bicolor</i> TURCZ. var. <i>japonica</i> NAKAI	Leguminosae	Sept. 29	1.332	0.392	1.724	77.2	
<i>Pueraria hirsuta</i> MATSUM.		"	" 30	1.122	0.501	1.626	69.0
<i>Desmodium racemosum</i> DC.		"	Oct. 9	1.261	0.427	1.688	74.7
<i>Vicia Faba</i> L. f. <i>ascendens</i> MAKINO (potted)	"	" 31	1.015	0.114	1.129	89.9	
<i>Polygonum Blumei</i> MEISN.	Polygonaceae	" 4	1.100	0.245	1.345	81.7	
<i>Polygonum Reynoutria</i> MAKINO		" 11	0.796	0.149	0.945	84.2	
<i>Rumex japonicus</i> MEISN.		" 4	0.916	0.109	1.025	89.3	
<i>Rumex Acetosella</i> L.	"	" 4	0.916	0.109	1.025	89.3	
<i>Clematis paniculata</i> THUNB.	Ranunculaceae	Nov. 22	0.503	0.101	0.604	83.2	
* <i>Cimicifuga acerina</i> TANAKA		Oct. 25	0.712	0.220	0.932	76.3	
<i>Thalictrum Thunbergii</i> A. P. DC. var. <i>hypoleucum</i> NAKAI		" 28	0.369	0.243	0.612	60.3	
<i>Amarantus Blitum</i> L. var. <i>oleraceus</i> HOOK. f.	Amarantaceae	Nov. 2	2.270	0.401	2.671	84.9	
<i>Celosia argentea</i> L.		Oct. 3	2.017	0.107	2.124	94.9	
"		" 12	1.210	0.148	1.358	89.1	

<i>Perilla frutescens</i> BRIT. var. <i>crispa</i> DECNE. f. <i>discolor</i> MAKINO	Labatae	Oct. 7	0.531	0.687	1.218	43.5
* <i>Salvia nipponica</i> MIQ. f. <i>argutidens</i> MAKINO	"	" 26	—	0.217	0.217	—
<i>Agrimonia japonica</i> KOIDZ.	Rosaceae	Sept. 28	1.714	0.051	1.765	97.1
<i>Chenopodium album</i> L. var. <i>centrorubrum</i> MAKINO	Chenopodiaceae	" 30	1.977	0.209	2.186	90.4
"	"	Oct. 2	1.942	0.154	2.096	92.6
<i>Cissus japonica</i> WILLD.	Vitaceae	" "	0.840	0.266	1.126	76.3
<i>Plantago major</i> L. var. <i>asiatica</i> DECNE. in the shade	Plantaginaceae	" 6	0.311	0.247	0.558	55.7
<i>Abutilon Arizense</i> GAERTN.	Malvaceae	" 7	1.650	0.137	1.787	92.3
<i>Metaplexis japonica</i> MAKINO	Asclepiadaceae	" "	3.404	0.112	3.516	96.8
<i>Calystegia japonica</i> CHOISY	Convolvulaceae	" 8	0.932	0.089	1.021	91.2
<i>Commelina communis</i> L.	Commelinaceae	" 12	0.844	0.145	0.989	85.3
<i>Humulus Lupulus</i> L. var. <i>cordifolius</i> MAXIM.	Moraceae	" "	0.770	0.267	1.037	74.2
<i>Astilbe congesta</i> NAKAI	Saxifragaceae	" 13	0.529	0.452	0.981	53.9
* <i>Phryma leptostachya</i> L.	Phrymaceae	" "	—	—	0.483	—
<i>Geranium nepalense</i> SWEET	Geraniaceae	" 15	0.880	0.720	1.600	55.0
<i>Impatiens Noli-tangere</i> L.	Balsaminaceae	" 16	—	—	0.849	—
* <i>Boehmeria japonica</i> MIQ.	Urticaceae	" 18	0.375	0.355	0.730	51.3
<i>Capsella Bursa-pastoris</i> MOENCH var. <i>auriculata</i> MAKINO	Cruciferae	" 21	1.177	0.078	1.255	93.7
<i>Oenanthe stolonifera</i> DC.	Umbelliferae	" 22	1.053	0.232	1.285	81.9
* <i>Begonia Evansiana</i> ANDR. (in the shade)	Begoniaceae	" 23	0.126	0.091	0.217	58.0
" (in the sun)	"	" "	0.266	0.261	0.527	50.4
<i>Sedum lineare</i> THUNB.	Crasulaceae	" "	0.295	0.042	0.337	87.5
<i>Phytolacca decandra</i> L.	Phytolaccaceae	" "	1.941	0.132	2.073	93.6
<i>Lilium Henryi</i> BAKER	Liliaceae	" 24	0.473	0.315	0.788	60.0
<i>Stellaria aquatica</i> SCOP.	Caryophyllaceae	" "	0.846	0.083	0.929	91.0
<i>Oenothera odorata</i> JACQ.	Oenotheraceae	" 26	0.856	0.122	0.978	87.5
<i>Crinum asiaticum</i> L. var. <i>japonicum</i> BAK.	Amaryllidaceae	" 30	0.459	0.041	0.500	91.8
<i>Trichosanthes japonica</i> REGEL	Cucurbitaceae	" "	0.970	0.184	1.154	84.0
<i>Papaver orientale</i> L.	Papaveraceae	Nov. 2	0.343	0.116	0.459	74.7
<i>Tropaeolum majus</i> L. (potted)	Tropaeolaceae	" "	2.603	0.521	3.124	83.3
<i>Oxalis corniculata</i> L.	Oxalidaceae	" 3	1.115	0.086	1.201	92.8
<i>Onoclea sensibilis</i> L.	Polypodiaceae	Oct. 20	0.382	0.562	0.944	40.4
* <i>Dryopteris viridescens</i> O. KUNTZE	"	" 31	0.482	0.143	0.625	77.1
<i>Phyllospadix Scouleri</i> Hook. marine	Polymogonaceae	Nov. 16	0.552	0.045	0.597	92.4

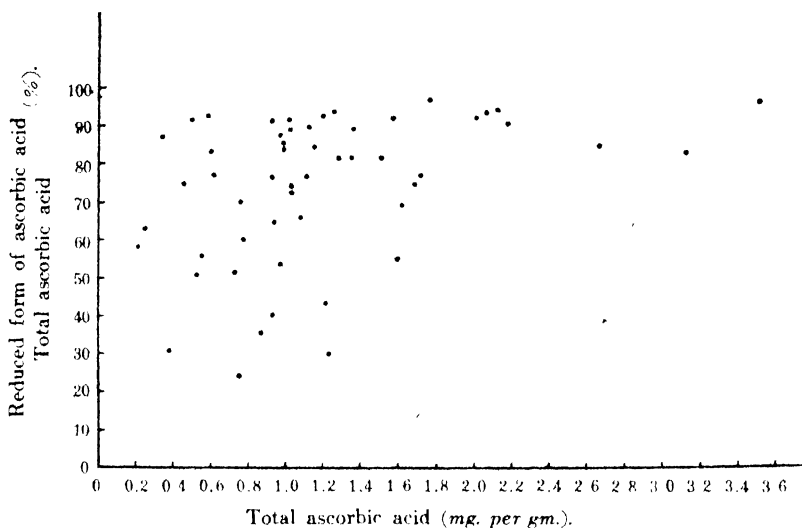


Fig. 1. Relation between reduced form of ascorbic acid/total ascorbic acid and total ascorbic acid.

The table shows also that herbaceous plants of small vitamin-C content belong, for the most part, to shade plants (mg. per gm.), viz.:

<i>Begonia Evansiana</i> ANDR.	(0.207)
<i>Salvia nipponica</i> MIQ. f. <i>argutidens</i> MAKINO	(0.217)
<i>Cacalia delphiniifolia</i> SIEB. et ZUCC.	(0.393)
<i>Cacalia bulbifera</i> MAXIM.	(0.446)
<i>Phryma leptostachya</i> L.	(0.483)
<i>Cimicifuga acerina</i> TANAKA	(0.618)
<i>Dryopteris viridescens</i> O. KUNTZE	(0.625)

the plant, grown by chance, in the shade, viz.:

<i>Plantago major</i> L. var. <i>asiatica</i> DECNE.	(0.558),
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the marine plant, viz.:

<i>Phyllospadix Scouleri</i> HOOK.	(0.597),
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the succulent leaves viz.:

<i>Crinum asiaticum</i> L. var. <i>japonicum</i> BAK.	(0.500)
<i>Sedum lineare</i> THUNB.	(0.337),

and *Papaver orientale* L. (0.459).

The herbaceous plants of large vitamin-C content mainly belong to sun plants. And among them, those of largest vitamin-C content are as follows (mg. per gm.):

<i>Metaplexis japonica</i> MAKINO	(3.516)
<i>Tropaeolum majus</i> L.	(3.124)
<i>Thalictrum Thunbergii</i> A. P. DC.	
var. <i>hypoleucum</i> NAKAI	(2.671)
<i>Chenopodium album</i> L.	
var. <i>centrorubrum</i> MAKINO	(2.186)
<i>Amarantus Blitum</i> L. var. <i>oleraceus</i> Hook. f.	(2.124)

From the data of MIWA ('38 IV, V, VII) herbaceous plants whose leaves have more than 1 mg. vitamin-C per gm. fresh weight are selected (only the reduced form was estimated):

Species	Family	Vitamin-C	Collection
<i>Sasa Veitchii</i> REHD.	<i>Gramineae</i>	1.421	Aug.
<i>Pleioblastus variegata</i> MAKINO var.			
<i>viridis</i> MAKINO, f. <i>pubescens</i> MAKINO	..	2.053-2.346	Feb.
<i>Hordeum sativum</i> JESS. var.			
<i>hexastichon</i> L.	..	0.897-1.477	May
<i>Agrostis perennans</i> TUCK.	..	1.079-1.177	Mar.
<i>Aster Laulureanus</i> FRANCH.	<i>Compositae</i>	1.111-1.228	Oct.
<i>Lactuca chinensis</i> MAKINO	..	1.079	Mar.
<i>Trifolium repens</i> L.	<i>Leguminosae</i>	1.295-1.523	Oct.
<i>Cassia Tora</i> L.	..	1.750-1.815	..
<i>Mimosa pudica</i> L.	..	1.372	Mar.-May
<i>Astragalus sinicus</i> L.	..	1.177	Feb.
<i>Pisum sativum</i> L.	..	0.875-1.027	Apr.
<i>Polygonum Blumei</i> MEISN.	<i>Polygonaceae</i>	1.556-1.729	Oct.
" <i>orientale</i> L.			
var. <i>pilosum</i> MEISN.	..	1.290-1.532	..
<i>Rumex Acetosella</i> L.	..	1.290	..
<i>Lobelia radicans</i> THUNB.	<i>Campanulaceae</i>	1.204-1.307	..
<i>Viola yezoensis</i> MAXIM.	<i>Violaceae</i>	1.156-1.103	..
" <i>mandshurica</i> W. BECK.			
var. <i>ciliata</i> NAKAI	..	1.406-1.758	Mar.-May
<i>Belamcanda chinensis</i> LEMAN	<i>Iridaceae</i>	2.751-3.075	Oct.
<i>Tritonia Pottii</i> BENTH. et HOOK. f.	..	1.313-1.688	Apr.
<i>Amarantus Blitum</i> L.			
var. <i>oleraceus</i> HOOK. f.	<i>Amarantaceae</i>	0.972-1.061	Oct.
<i>Alpinia japonica</i> MIQ.	<i>Zingiberaceae</i>	1.045-1.088	..
<i>Aspidistra elatior</i> BLUME	<i>Liliaceae</i>	1.428-1.493	Feb.
<i>Agapanthus umbellatus</i> L'HER.	..	1.181-1.313	Apr.
<i>Galium trifidum</i> L.	<i>Rubiaceae</i>	1.233	Mar.
<i>Potentilla Freyniana</i> BORNH.	<i>Rosaceae</i>	1.126-1.177	..
<i>Carex Prescottiana</i> BOOTT			
var. <i>kiotensis</i> KUEK.	<i>Cyperaceae</i>	0.989-1.148	May
<i>Capsella Bursa-pastoris</i> MOENCH.			
var. <i>auriculata</i> MAKINO	<i>Cruciferae</i>	1.083	Mar.
<i>Veronica peregrina</i> L.	<i>Scrophulariaceae</i>	1.126	Feb.
<i>Microlepia strigosa</i> PRESL	<i>Polypodiaceae</i>	0.967-1.128	Mar.

Similarly from the data of SAITO and WATANABE ('39) we get as follows :

Species	Family	Vitamin-C	Collection
<i>Melilotus suaveolens</i> LEDERBOUR	<i>Leguminosae</i>	1.2963	Aug.
<i>Chenopodium centrorubrum</i> NAKAI	<i>Polygonaceae</i>	1.2144	..
<i>Persicaria Bungeana</i> NAKAI	..	2.8478	..
<i>Persicaria nodosa</i> OPIZ.	..	1.7118	..
<i>Persicaria lapathifolia</i> S. F. GRAY	..	1.1621	..
<i>Rumex callosus</i> RECHINGER FIL.	..	1.1667	..
<i>Persicaria cochinchinensis</i> KITAGAWA	..	1.3088	..
<i>Leonurus sibiricus</i> L.	<i>Labiatae</i>	1.1842	..
<i>Amarantus retroflexus</i> L.	<i>Amarantaceae</i>	1.0599	..
<i>Abutilon Avicennae</i> GAERTNER	<i>Malvaceae</i>	1.1542	..

As the redox-system, according to BAUR's scheme ('32-'37) makes a cycle, viz., from ascorbic acid to dehydroascorbic acid and from dehydroascorbic acid back to ascorbic acid, such a small amount of ascorbic acid as exists in each green plant is sufficient for photosynthesis (BUKATSCH '39).

The shade plants have indeed a somewhat lower content of chlorophyll than the sun plants per unit surface of the leaf, but higher, per fresh weight - that is, the chlorophyll factor in the shade plants is therefore sufficient for photosynthesis (LUNDEGÅRDH '22). The shade plants, however, are found to assimilate CO₂ in a smaller amount than the sun plants under natural conditions (HIRAMATSU '32). This fact may be a cause of the presence of a smaller content of ascorbic acid in shade plants, for, in the sun, as long as they are in a normal condition, the shade plants have a higher assimilatory activity (see HIRAMATSU '32) and higher content of vitamin-C than in the shade (Table I, *Begonia Evansiana* ANDR.) and the sun plants have a lower content of vitamin-C in the dark than in the light (Table II). We shall discuss this subject further in the next chapter. That the shade plant seems to have relatively large quantities of dehydroascorbic acid, may have some meaning with regard to the necessity of both forms of ascorbic acid in BAUR's scheme; it is, however, necessary to estimate them more exactly (O₂ being removed and ascorbic acid oxidase, inactivated) and systematically especially in the case of low vitamin-C and high ascorbic acid oxidase content, in which we often meet with rapid oxidation.

A lower vitamin-C content of succulent leaves -- we have only one or two examples -- may be explained by other factors than the light condition, for instance, a larger water content (*Sedum lineare* THUNB.

96% ; *Crinum asiaticum* L. var. *japonicum* BAK. 90%): Because most succulent leaves which have smaller surface area, seem, indeed, to be at a disadvantage in utilizing sun light, but their internal structure compensates for it (SCHANDERL '35).

2. *The effect of light on the accumulation of ascorbic acid in leaves.*

As stated above, the shade plant in the sun, so long as it remains in a normal condition, has a higher content of vitamin-C than in the shade (Table I, *Begonia Evansiana* ANDR.) and the sun plant has a lower vitamin-C content in the dark and is restored to its original value under illumination (Table II). This provides an illustration of the effect of

TABLE II.

Effect of Light on Accumulation of Ascorbic Acid in Leaves of Vicia Faba L. f. ascendens MAKINO (potted).

Date (collect. and exp.)	Ascorbic acid content			Remarks
	Reduced form	Oxidized form	Total	
(1939)	mg. per gm.	mg. per gm.	mg. per gm.	
Oct. 31, 3.00 p.m.	1.015	0.114	1.129	placed in the dark room at 5.30 p.m.
Nov. 1, 2.20 p.m.	0.896	0.002	0.898	darkness
" 2, 1.50 p.m.	0.817	0.047	0.864	darkness
" 4, 11.00 a.m.	0.725	0.023	0.748	exposed to the light at 1.20 p.m.
" 5, 10.00 a.m.	0.918	0.015	0.933	light
" 6, 10.00 a.m.	1.019	0.049	1.068	light

light upon the vitamin-C synthesis. In this experiment potted horse-bean plants, about a month old, were used. The vitamin-C content of their leaves was estimated both in the dark and under illumination. As the light source a Mazda lamp of 200 W. with paraboloidal chrome-plated reflector was used, from which light passes through a thick layer of water to plants at a distance of 40 cm. The extraction of vitamin-C of the leaves in the darkness was carried out in the darkness.

It was generally accepted that light has a remarkable effect on the synthesis of the ascorbic acid during germination, though in seedlings grown even in darkness, the ascorbic acid accumulates for several days (HELLER '28, RUBIN and STRACHIZKY '36 etc.). It is not yet known whether the loss of ascorbic acid in the darkness is a result of destruction or utilization or both of the substance without new formation, due to the

TABLE

Ascorbic Acid Content of Leaves of Several Varieties of

Date (collect. and exp.)	Locality Ascorbic acid content	Asahikawa (Hokkaidō) 43° 47' N. Lat.			Remarks	Sendai	
		Reduced form	Oxidized form	Total		Reduced form	Oxidized form
(1939)		mg. per gm.	mg. per gm.	mg. per gm.		mg. per gm.	mg. per gm.
Oct. 8		0.966	0.193	1.159	matured seeds in scattering; plant in a vital state.	0.644	0.515
.. 19		0.853	0.327	1.180	seeds have scattered; plant in a reduced state.	0.602	0.339
Nov. 6		1.040	0.118	1.158	upper stem withered; plant in a reduced state; leaves speckled with anthocyanin.	0.868	0.253
.. 24		0.995	0.114	1.090	water content: 69.1% leaves speckled with anthocyanin; plant in withering	0.563	0.087

TABLE

Ascorbic Acid Content of Leaves of Several Varieties of Desmodium

Date (collect. and exp.)	Locality Ascorbic acid content	Yoiti (Hokkaidō) 43° 12' N. Lat.			Remarks	Sendai	
		Reduced form	Oxidized form	Total		Reduced form	Oxidized form
(1939)		mg. per gm.	mg. per gm.	mg. per gm.		mg. per gm.	mg. per gm.
Oct. 9		0.957	0.368	1.325	seeds have fallen.	1.261	0.427
.. 29		0.542	0.382	0.924	greater part of leaves have not yet fallen.	0.577	0.325
Nov. 22							
.. 25							

For the materials used in these experiments, the authors are indebted to

III.

Miscanthus sinensis ANDERS. grown in Same Soil at Sendai.

(Miyagi-prefecture) 38° 15.5' N. Lat.			Kōti (Kōti-prefecture) 33° 33' N. Lat.		
Total	Remarks	Reduced form	Oxidized form	Total	Remarks
mg. per gm.		mg. per gm.	mg. per gm.	mg. per gm.	
1.159	in flowering.	0.579	0.353	0.932	about to be in the ear.
0.941	flowers being over; the ears being with feathers; plant in a vital state.	0.527	0.379	0.906	beginning to flower; plant in a vital state.
1.121	seeds in scattering; plant in a vital state. water content: 66.8%.	0.818	0.247	1.065	in flowering; plant in a vital state. water content: 67.5%.
0.650	leaves speckled with anthocyanin; plant in withering.	1.301	0.310	1.611	flowers being over; plant in a vital state.

IV.

racemosum DC. grown in Botanical Garden of Biological Institute.

(Miyagi-prefecture) 38° 15.5' N. Lat.			Misaki (Kanagawa-prefecture) 35° 08' N. Lat.		
Total	Remarks	Reduced form	Oxidized form	Total	Remarks
mg. per gm.		mg. per gm.	mg. per gm.	mg. per gm.	
1.688	seeds on the point of falling; yellowish tint of leaves being deeper than in that from Yōiti.	0.642	0.493	1.135	plant being larger; in flowering;
0.902	leaves begin to fall; leaves as green as possible were used for analysis.	0.570	0.439	0.909	flowers being almost over; plant in a vital state.
		0.422	0.141	0.563	leaves being of yellowish tint and on the point of falling; greater part of seeds have not yet fallen.
		0.402	0.161	0.563	leaves being of yellowish tint and on the point of falling.

Dr. Y. NAKAJIMA of the Biological Institute.

influence of light on the activity of the chloroplasts. It seems, however, certain that a given concentration of ascorbic acid in grown-up leaves corresponds, within the physiological limit, to a given intensity of light; that is, when the intensity of light increases up to a certain value, the concentration of ascorbic acid increases and a new equilibrium is attained. This was the case in young plants (SUGAWARA '39, REID '38). Some of the ascorbic acid synthesized in the leaves of seedlings in light is transported to other organs, and some of the carbohydrates synthesized by the leaves are transported there and are converted into ascorbic acid; the ascorbic acid accumulates rapidly during the period of the expansion of the leaf, and when full size is attained there usually is no further increase apparent unless the light or nutritive conditions of the plants are changed. It is possible that they, however, continue the synthesis and translocate or in some way utilize the excess (REID '38).

3. Variety study.

Variety is considered to be a factor of considerable importance in determining the ascorbic acid of vegetables, fruits etc. (TRESSLER, MACK and KING '36).

A general résumé of our preliminary studies is presented in Tables III and IV. From these data it is seen that (1) there was not much difference in the ascorbic acid content of varieties of *Miscanthus sinensis* ANDERS. and a little difference in the case of varieties of *Desmodium racemosum* DC. at the active stages of maturity, at which these plants were examined and (2) in reduced state of leaves ascorbic acids decreased.

As these variety are those which had adapted themselves to different climates, the plants could not, of course, be compared with each other at the same stage of maturity at the same time.

4. The vitamin-C content of marine algae.

The vitamin-C content of marine algae has been determined by many authors (NORRIS, SIMEON and WILLIAMS '37, LUNDE and LIE '38 etc. see also KING '39). According to NORRIS *et al.* ('37) the marine algae at the coast have, generally, a higher vitamin-C content than those at the sea-bottom, which fact may be to a certain extent due to the difference of the light intensity at their habitats as in the case of herbaceous plants.

The present materials are limited to the habitats of the coastal zone near the Marine Biological Station, Asamushi at which this experiment was carried out during November 1939. Extraction and analysis were made similarly except that the materials were not washed with distilled water but with filtered sea-water.

The results are summarized in Table V.

TABLE V.
Vitamin-C Content of Marine Algae.

Material*	Class	Date (collect. and exp.)	Vitamin-C content		
			Reduced form	Oxidized form	Total
<i>Chondria crassicaulis</i> HARV. (old)	<i>Rhodophyceae</i>	(1939) Nov. 16	0.473	0.061	0.534
" " young	"	" "	0.191		0.191
<i>Hypnea seticulosa</i> J. Ag.	"	" "	0.098	0.107	0.205
<i>Rhodomela Larix</i> (TURN.) C. Ag	"	" 17	-	-	0.046
<i>Coccophora Langsdorfi</i> (TURN.) GREV. (in its 1st year)	<i>Phaeophyceae</i>	" 13	0.783	0.156	0.939
<i>Coccophora Langsdorfi</i> (TURN.) GREV. (in its 2nd year)	"	" "	0.704	0.073	0.777
<i>Chorda Filum</i> (L.) LAMOUR.	"	" 14	0.980		0.980
<i>Sargassum tortile</i> C. Ag.	"	" 17	1.879	1.438	3.317
<i>Enteromorpha compressa</i> (L.) GREV.	<i>Chlorophyceae</i>	" 14	0.156	0.100	0.256
<i>Ulva pertusa</i> KJELLM.	"	" "	0.256	0.154	0.410
<i>Chaetomorpha moniligera</i> KJELLM.	"	" 16	0.294	0.103	0.397

* Authors are grateful to Drs. M. TAKAMATSU and K. ABE for the identification.

SUMMARY

1. The vitamin-C content of wild herbaceous plants was estimated.
2. Generally, those of high vitamin-C content belong to the sun plant and those of low content to the shade plant. This fact seems to be correlated with assimilatory activity in the sense of BAUR's scheme.
3. The greater parts of vitamin-C of herbaceous plants are found in reduced form.
4. The relatively higher content of oxidized form in the shade plant than in the sun plant may have some meaning in photosynthesis. A further exact analysis, however, is necessary.
5. The ascorbic acid decreases in darkness and is restored in light. The shade plant in the sun contains a higher vitamin-C than in the shade, within the limit of its survival. That is, the vitamin-C content of the leaves is due to the intensity of light.
6. The vitamin-C content of leaves of varieties of *Miscanthus sinensis* ANDERS. and *Desmodium racemosum* DC., which had been adapted to

different climates was estimated. In the varieties of the former, there was not much difference and in the latter, a little.

7. The vitamin-C content of marine algae was determined.

REFERENCES

- ASIKAGA, M. 1938. Studies about the ascorbic acid in sugar cane. (*Japanese*). *J. Soc. Trop. Agr.*, **10**, 412.
- BAUR, E. 1932. Desensibilatoren, Antioxygene und Antifluorescenten. *Z. physik. Chem.*, **16**, 465.
- . 1935. Über eine photochemische Reaktion von Chlorophyll. *Helvet. chim. Acta*, **18**, 1157.
- . 1937. *a.* Über den Verlauf der Photolyse der Kohlensäure. *ibid.*, **20**, 387.
- und FRICKER, H. 1937. *b.* Über die photolytische Bildung von Formaldehyd aus Chlorophyll und Eosin. *ibid.*, **20**, 391.
- und GLOOR, K. 1937. *c.* Über die photolytische Bildung von Formaldehyd in der Eosingruppe. *ibid.*, **20**, 970.
- BESSEY, O. A. and KING, C. G. 1933. The distribution of vitamin C in plant and animal tissue and its determination. *J. biol. Chem.*, **103**, 687.
- . 1938. *a.* A method for the determination of small quantities of ascorbic acid and dehydroascorbic acid in turbid and colored solution in the presence of other reducing substances. *J. biol. Chem.*, **126**, 771.
- . 1938. *b.* Vitamin C. Methods of assay and dietary sources. *J. Amer. Med. Ass.*, **111**, 1290.
- BORSOOK, H., DAVENPORT, H. W., JEFFEREYS, C. E. P. and WARNER, R. C. 1937. The oxidation of ascorbic acid and its reduction in vitro and in vivo. *J. biol. Chem.*, **117**, 237.
- BUKATSCH, F. 1939. Über die Rolle der Ascorbinsäure in den Chloroplasten. *Planta*, **30**, 118.
- CARUSO, C. 1938. Nuove osservazioni sulla localizzazione di talune reazioni argentiche. *Protopl. (Berl.)*, **31**, 489.
- DISCHENDORFER, O. 1937. Über den histochemischen Nachweis von Vitamin C (l-Ascorbinsäure) in Pflanzen. *Protopl. (Berl.)*, **28**, 516.
- FOX, F. S. and LEVY, L. F. 1936. Experiments confirming the antiscorbutic activity of dehydroascorbic acid and a study of its storage and that of ascorbic acid by the guinea-pig at different levels of intake. *Bioch. J.*, **30**, 221.
- FUJITA, A. und EBIHARA, T. 1937. Über die Verteilung des Vitamin C in tierischen und pflanzlichen Geweben. 1. *Bioch. Z.*, **290**, 201.
- und ———. 1939. *a.* II. *ibid.*, **300**, 143.
- und ———. 1939. *b.* Über die gebundene Ascorbinsäure in pflanzlichen Geweben. *ibid.*, **301**, 229.
- GIROUD, A., LEBLOND, C. P. et RATSIMAMANGA, R. 1934. Signification de la reduction des sels d'argent au niveau des plastes chlorophylliens. *C. r. Soc. Biol., Paris*, **117**, 614.
- , RATSIMAMANGA, R. et LEBLOND, C. P. 1934. Parallélisme entre la vitamine C et la chlorophylle. *ibid.*, **117**, 612.
- . 1938. L'acide ascorbique dans la cellule et les tissus. *Protopl. (Berl.)*, *Monogr.*, **16**.

- HELLER, V. G. 1928. Vitamin synthesis in plants as affected by light source. *J. biol. Chem.*, **76**, 499.
- HIRAMATSU, K. 1932. On the daily progress of carbon assimilation in the shadow under natural conditions. *Sci. Rep. Tôhoku Imp. Univ., Biol.*, **7**, 239.
- HIRST, E. L. and ZILVA, S. S. 1933. Ascorbic acid as the antiscorbutic factor. *Bioch. J.*, **27**, 1271.
- IJIDO, J. B. H. 1936. The influence of fertilizers on the carotene and vitamin C content of plants. *Bioch. J.*, **30**, 2307.
- ITO, N. 1938. Studies on ascorbic acid oxidase. 1. (*Japanese with English abstract*). *Bull. Agr. Chem. Soc. Japan*, **14**, 140.
- KATAI, K. 1939. On the determination of ascorbic acid and the presence of combined ascorbic acid. (*Japanese*). *Bull. Sci. Fakul. Terkult. Kyusyu Imp. Univ. Hukuoka, Japan*, **8**, 286.
- KING, C. G. 1939. The water-soluble vitamins. Vitamin C (Ascorbic acid). *Ann. Rev. Bioch.*, **8**, 392.
- LUNDEGÅRDH, H. 1922. Zur Physiologie und Ökologie der Kohlensäureassimilation. *Biol. Zent.*, **42**, 337.
- LUNDE, G. and LIE, J. 1938. Vitamin C in Meeresalgen. *Z. physiol. Chem.*, **254**, 227.
- MCHEHRY, E. W. and GRAHAM, M. 1935. Observations on the estimation of ascorbic acid by titration. *Bioch. J.*, **29**, 2013.
- MENAKER, M. H. and GUERRANT, N. B. 1938. Standardization of 2, 6-dichlorophenolindophenol. An improved method. *Ind. Eng. Chem., Anal. Ed.*, **10**, 25.
- MIRIMANOFF, A. 1938. Acide ascorbique et pigments caroténoides. Signification de la réaction de MOLISCH et essai de localisation de l'acide ascorbique. *C. r. Acad. Sci. Paris*, **206**, 1038.
- . 1939. a. Le dosage et la localisation de la vitamin C dans les tissus végétaux. *Rev. Cytol. et Cytophysiol. végét.*, **3**, 119.
- . 1939. b. Remarques sur la réduction du nitrate d'argent au niveau des chloroplastes. *Rev. gén. Bot.*, **51**, 133.
- MITUDA, H. 1938. a. Studies on vitamin C. I. On the vitamin C content in various flowers and leaves. (*Japanese with English abstract*). *Bull. Agr. Chem. Soc. Japan*, **14**, 1228.
- . 1938. b. II. On the variation of vitamin C content in some fruits. (*Japanese with English abstract*). *ibid.*, **14**, 1335.
- MIWA, H. 1938. Studies on vitamin-C resources. I. The vitamin-C content of leaves of trees in late autumn. (*Japanese*). *Jôzôgaku-Zasshi*, **16**, 581.
- . 1938. II. The vitamin-C content of young leaves of trees in early summer. (*Japanese*). *ibid.*, **16**, 716.
- . 1938. III. On the vitamin-C content of wild herbaceous plants. (*Japanese*). *ibid.*, **16**, 798.
- . 1938. IV. The vitamin-C content of leaves of trees in summer. (*Japanese*). *ibid.*, **16**, 937.
- . 1939. V. On the vitamin-C content of wild herbaceous plants (August—October). (*Japanese*). *ibid.*, **17**, 33.
- . 1939. VI. On the vitamin-C content of leaves of trees (September—October). (*Japanese*). *ibid.*, **17**, 177.
- . 1939. VII. (*Japanese*). *ibid.*, **17**, 579.
- NORRIS, E. R., SIMEON, M. K. and WILLIAMS, H. B. 1937. The vitamin B and vitamin C

- content of marine algae. *J. Nutrit.*, **13**, 425.
- OKUDA, Y. and KATAI, K. 1939. Biochemical investigation of mosaic disease of tobacco plants V. On the ascorbic acid content in the leaves of healthy and mosaic plants (Japanese). *Bull. Agr. Chem. Soc. Japan*, **15**, 80.
- PEKAREK, J. 1938. Die Lokalisation des Silbernitrat-Reduktors in den Chloroplasten. *Protopl. (Berl.)*, **30**, 534.
- RANDOIN, L., GIROUD, A. et LEBLOND, C. P. 1935. Recherches biologiques et biochimiques sur la teneur en acide ascorbique des tissus chlorophylliens et achlorophylliens. *Bull. Soc. Chim. Biol. Paris*, **17**, 1649.
- REEDMAN, E. J. and MCHENRY, E. W. 1938. Combined ascorbic acid in plant tissues. *Bioch. J.*, **32**, 85.
- REID, M. E. 1938. The effect of light on the accumulation of ascorbic acid in young cowpea plants. *Amer. J. Bot.*, **25**, 701.
- RUBIN, B. A. and STRACHIZKY, K. 1936. On the biological role of vitamin C in the plant. *Biokhimiia (Biochimia)*, **1**, 343.
- SAITO, M. and WATANABE, M. 1939. Studies on wild grasses of Manchoukuo as fodder plants. I. (Japanese). *Rep. Inst. Sci. Res. Manchoukuo*, **3**, 39.
- SANSOME, F. M. and ZILVA, S. S. 1936. Polyploidy and vitamin C. *Bioch. J.*, **30**, 54.
- SCHANDERL, H. 1935. Untersuchungen über die Lichtverhältnisse im innern von Hartlaub- und Sukkulantenblättern. *Planta*, **24**, 454.
- SUGAWARA, T. 1939. Studies on the formation of ascorbic acid (vitamin C) in plants. I. The influence of light on the ascorbic acid contents in various etiolated seedlings. *Jap. J. Bot.*, **10**, 141.
- , —, 1938. Influence of fertilizer on the ascorbic acid content of spinach. (Japanese with English summary). *J. Sci. Soil and Manure, Japan*, **12**, 567.
- TILMANS, J. 1930. Das antiskorbutische Vitamin. *Z. Unters. Lebensm.*, **60**, 34.
- , —, HIRSCH, P. und HIRSCH, W. 1932. Das Reduktionsvermögen pflanzlicher Lebensmittel und seine Beziehung zum Vitamin C. I. Der reduzierende Stoff des Citronensaftes. *ibid.*, **63**, 1.
- , —, und SIFFERT, F. 1932. II. Über die Frage, ob der reduzierende Stoff des Citronensaftes als Stabilisator für das eigentliche Vitamin fungiert. *ibid.*, **63**, 21.
- , —, und JACKISCH, J. 1932. III. Der Gehalt der verschiedenen Obst- und Gemüsearten an reduzierendem Stoff. *ibid.*, **63**, 241.
- , —, und DICK, H. 1932. IV. Über die Reversibilität der Oxydationen des reduzierenden Stoffes im Citronensaft. *ibid.*, **63**, 267.
- , —, und JACKISCH, J. 1932. V. Die antiskorbutische Wirkung verschiedener Auszüge der Gurke. *ibid.*, **63**, 276.
- TRESSLER, D. K. and MACK, G. L. and KING, C. G. 1936. Factors influencing the vitamin C content of vegetables. *Amer. J. publ. Health and nat. Health*, **26**, 905.
- WEIER, E. 1938. Factors affecting the reduction of silver nitrate by chloroplasts. *Amer. J. Bot.*, **25**, 501.

STUDIES OF CLEAVAGE I. CHANGES IN SURFACE AREA OF DIFFERENT REGIONS OF EGGS OF A SEA URCHIN IN THE COURSE OF THE FIRST CLEAVAGE*

By

ISAO MOTOMURA

Biological Institute, Tôhoku Imperial University, Sendai

(With 5 text-figures)

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It is a well known fact that the position of the polarity axis of the egg of the sea urchin is determined prior to the fertilization. As to the mechanism of this fact the author has developed the conception that in the cortical cytoplasm there exists a structure necessary for the determination (MOTOMURA '33, '35, '36). The hypothesis given here is substantiated by the following facts; 1) the cortical cytoplasm is so tough in contrast with the endoplasm that the former is not easily displaced by centrifugal force; 2) the cortical cytoplasm is not carried inside the embryo by the protoplasmic flow in the course of the normal cleavage; 3) the cell wall between the blastomeres of the normal egg is newly produced. Those facts, which were confirmed by the present author from observations on the behavior of the cortical cytoplasm of the egg of *Strongylocentrotus pulcherrimus*, contradict either the surface tension theory of cleavage advocated by SPEK ('18) or the theories which attribute cleavage to the mechanical pressure exerted by the hyaline layer or by the growing aster.

Recently SCHECHTMAN ('37) found the local extension of the cortical layer of the furrow region in the cleaving egg of amphibia. According to him the cleavage plane of the amphibian egg must be a part of the old cortical cytoplasm. In the sea urchin egg, K. DAN, J. C. DAN and T. YANAGITA ('38) concluded from observations on the movement of the surface of the cleaving egg that a new formation of surface takes place in the furrow region after cleavage has been completed, and further that the contact area between the blastomeres in normal eggs is composed of this newly formed surface. Now, the problem whether the cleavage plane is covered by the extension of the old cell surface or by a newly

* Contribution from the Marine Biological Station, Asamushi, Aomori-Ken, No. 166.

formed one must be an important one with respect to a theory of determination. In this paper, the results of observation on the cleavage of a sea urchin will be reported.

MATERIAL AND METHODS

The material used was the egg of a sea urchin, *Temnopleurus hardwickii*, at Asamusi. The fertilization membrane was removed by shaking the eggs in a test tube with sea water three minutes after fertilization. The eggs were then put into kaolin suspension in sea water and, after repeated washing, they were enclosed in a depression slide. Observation of the behavior of marks of the kaolin particles adhering to the surface of the egg was carried out from the beginning of the first cleavage till the appearance of the second cleavage furrow.

In order to know the relative amount of extension and contraction of the cortical cytoplasm, the surface areas of different regions of the eggs were measured. Assuming the geometrical form of the cleaving egg to be a revolution of the largest optical section in the side view of the egg, and the cleavage axis to be the revolution axis, the surface area was graphically measured in approximation as a summation of many sections of cones having a common revolution axis. Practically, measurements were carried out as follows (Fig. 1).

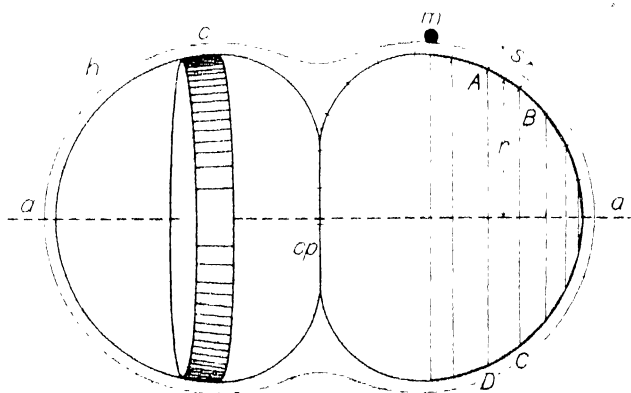


Fig. 1. Schematic representation of the method of measurement of the surface area of the egg in the first cleavage. a; revolution axis or cleavage axis. c; section of a cone. cp; cleavage plane. h; hyaline membrane. m; kaolin particle. r; radius of cone. s; lateral side 5 mm length.

When the bipolar spindle of the first cleavage appeared, the egg with its cleavage axis parallel to the microscopic field was sketched with

camera lucida at a magnification of 500 times. The position of the kaolin particles was also noted. The drawing was repeatedly continued every minute till the appearance of the second cleavage. The revolution axis was determined on the drawing by connecting the middle points of the respective diameters of the two blastomeres. The circumference of the drawing was scaled every 5 mm in straight distance beginning from the pole to the equator of the cleaving egg. By connecting these points the largest optical section of the egg is inscribed by a polygon, which has sides 5 mm length. The surface area of a revolution of this polygon around the cleavage axis is calculated with the equation

$$\text{area} = 2\pi(r_1s_1 + r_2s_2 + r_3s_3 + \dots + r_ns_n)$$

where r is the length of the perpendicular line from the middle point of the lateral side of a trapezoid ABCD, which is one of the sections of the polygon, to the revolution axis. And since, as mentioned above, the sides of the trapezoids are equally 5 mm in length, except the last one, the equation is modified to

$$\text{area} = 2\pi s \Delta r + 2\pi s_n r_n$$

where the last term is the lateral surface area of the last cone.

The surface area of the revolution of the inscribing polygon must be smaller than that of the original revolution. In the present authors cases the shortage came to 5 per cent. But this is not a great obstacle to determining the relative amount of the chronological changes, because the shortage is nearly equal in all cases providing the same method and magnification are employed. The probable error of measurement, which was caused mainly by the inexactness of the drawings, was less than 4 per cent.

OBSERVATIONS

1. Cases marked on the polar region.

In cases when marks of the kaolin particles were attached to the polar region of the egg, the area of the distal portion, which was less than 37 per cent of the half of the total surface area of the uncleaved egg, increased at the beginning of the cleavage, proceeded to a maximum, and then decreased gradually. The changes of the distal and the corresponding proximal areas in the course of the cleavage are given in Fig. 2. The expected value of the increase of the surface area must be about 26 per cent; that is, in total, 126 per cent of the initial area, assuming that the surface material of the uncleaved spherical egg covers

the total surface of the spherical blastomeres at the two cell stage without adding any new material to the cell surface. The observed value of the maximum of the distal portion exceeded this value and, on the other hand, the final value of this portion was always smaller than the expected value. On the contrary, the form of curves of the surface areas of the

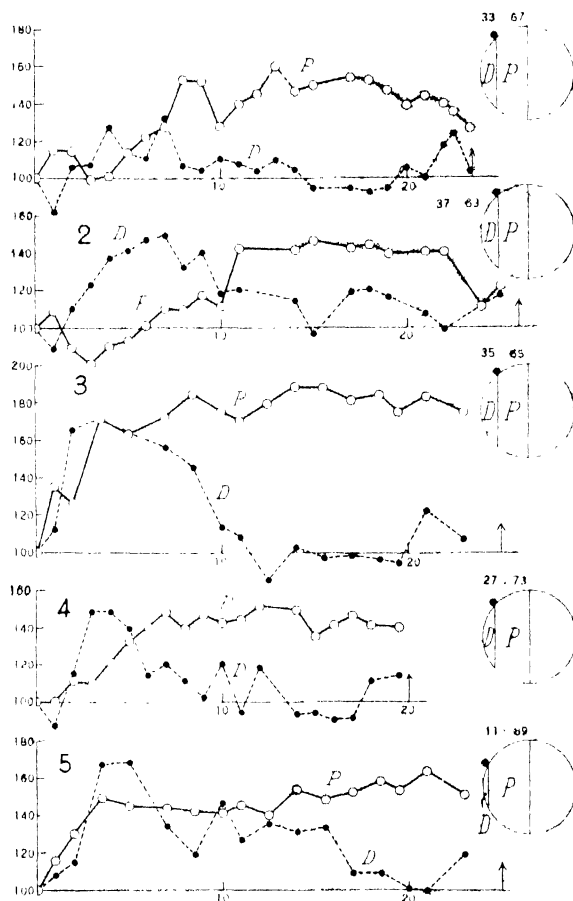


Fig. 2. Changes of the surface area of regions of the eggs in the first cleavage. Cases marked on the polar region, showing the initial extension and later contraction of the distal portions (D) and the increase of the proximal area (P) in the later half of the cleavage. Position of the kaolin particle is represented by ratio of areas of the distal and proximal portions in percentage of a half of the total surface area of the egg before cleavage. The areas of the portions at the beginning were taken as 100. Points more than 100 in the ordinates indicate increase. Abcissae, time in minutes.
 † Appearance of the second cleavage furrow.

corresponding proximal portions showed a slow increase at the start, and after one fourth of the cleavage time had elapsed, it showed a sudden increase, and then passed into a stationary state, in which the areas of the proximal portions were more than 126 per cent of the initial areas.

The results showed the same tendency in all observed cases. In company with the advance of the cleavage furrow, the increase of the surface area appeared at first at the distal region of the egg, and after the cleavage furrow had apparently been completed, the distal region contracted slowly, and the expansion of the proximal region began.

2. Cases having nearly equal area of the distal and proximal portions.

Two examples of these cases are given in Fig. 3. They showed the same tendency in regard to changes of the area as in the above mentioned

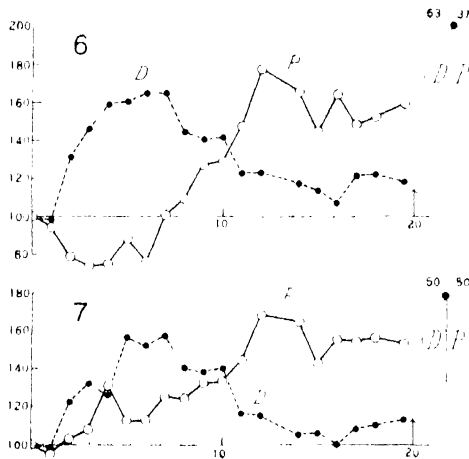


Fig. 3. Changes of the surface areas of proximal and distal regions of nearly equal initial size in the course of the first cleavage. Distal regions extended at the start and then contracted. Areas of the corresponding proximal regions increase in the later half over the expected value, 126 per cent.

cases. The area of the distal portion increased at the start of the cleavage and decreased after the completion of the cleavage furrow. The value of the contraction stage was less than 126 per cent of the initial area.

The proximal portion showed only a slight change at the start, and then suddenly extended over 126 per cent of the initial area, and in the later half of the cleavage time it became stationary.

The increase and decrease of the proximal and distal portions of these cases were delayed in comparison with the first cases. As a result,

the curves of the proximal and distal areas crossed at the middle of the total cleavage time.

3. Cases marked on the furrow region.

Three examples, No. 8, No. 9 and No. 10, in which the kaolin marks were put on the furrow region are described here (Figs. 4 and 5). The areas of the distal portions were 89, 74 and 96 per cent respectively. In those cases, the form of the curves of the distal portions looked like that of the total area, in which only a slight increase was visible at the start. After half of the cleavage time had elapsed, the area decreased gradually and nearly reached the initial amount.

The proximal portion showed, on the other hand, a very remarkable increase, which began after the constriction of the furrow was completed and reached a maximum when two thirds of the total cleavage time had passed. In example No. 10, the amount of increase reached 700 per cent

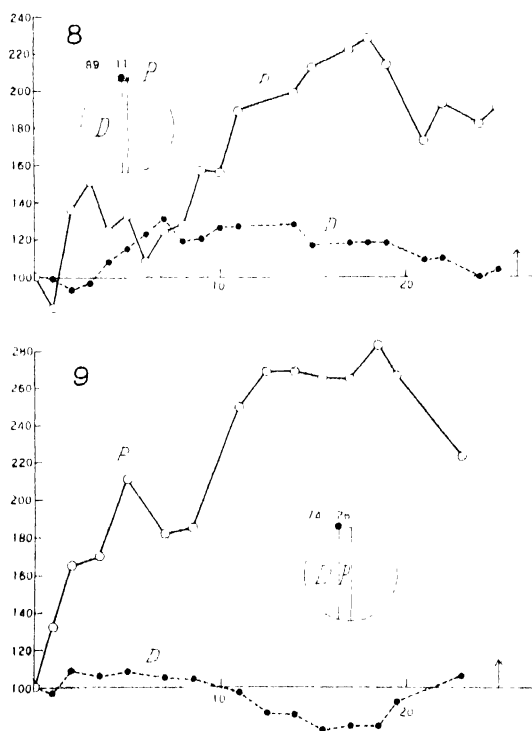


Fig. 4. Changes of the surface areas of regions of the cells marked on the furrow region in the course of the first cleavage. Increase of the proximal area is remarkable in comparison with that of the distal area.

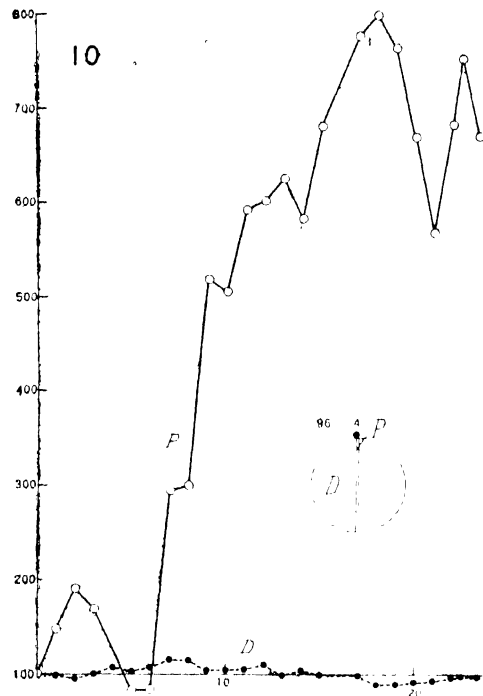


Fig. 5. Changes of the surface areas of regions of the cells marked on the furrow region in the course of the first cleavage. Notice that the increase of the proximal area was remarkable while the distal area remained nearly constant.

of the initial area. In other words, a small proximal portion, which was 4 per cent of half the total area of the uncleaved egg, increased to seven times, that is, 28 per cent. And the real increase amounts to $28 - 4 = 24$ per cent. This fact shows that most of the increase of the surface area in the last half of the cleavage time span is borne by the furrow region.

DISCUSSION

The results of measurement of the surface area of different regions of the egg of a sea urchin at the cleavage stage suggest many remarkable facts. In the first place, three periods are distinguishable in the time from the first appearance of the first cleavage furrow to the beginning of the second cleavage furrow. The first one third of that time is the constriction period, during which the cleavage furrow is apparently completed, forming a delicate protoplasmic bridge between two blastomeres.

In this period the extension of the surface of the egg is remarkable, especially at the pole. The second period, which is the second one third of the time, is characterized both by the contraction of the distal region and by the increase of the proximal area. The contraction of the distal region is so remarkable that in some cases the area of the distal portion is reduced to the initial area. The increase of area in the proximal region is greatest in the furrow region. In the last one third, the third period, the form of the curve is rather stationary. In this time the second cleavage is prepared.

DAN and others ('37 and '38) confirmed, from the measurement of changes of the arc length of optical sections of a sea urchin's egg in normal cleavage, that the maximum value of the curves become increasingly greater in the order, polar region, subpolar region, subfurrow region, and furrow region, and that the attainment of the maximum is delayed in the same order. It is very remarkable fact that the present author's results agree with theirs in many points, irrespective of the fact that the arc length is not proportionate to the surface area. The extension of the polar region at the beginning was pointed out by DAN. As mentioned above, the amount of extension exceeded the expected value. This fact shows that the extension of the polar region is not proportional to the increase of the total surface area but, on the other hand, that it is superior to the other regions of the egg. Next, the contraction of the polar region appears. As the result of the contraction the area of the distal portion again approaches the initial area. And the last phenomenon is always accompanied by a vigorous increase of area at the furrow region, by which the contraction of the distal portion is cancelled. It must be a most remarkable fact that, in the later periods of cleavage, the increase of the surface area is restricted only to a very limited furrow region.

In the egg of *Strongylocentrotus pulcherrimus*, orange red pigment granules are distributed in the cortical cytoplasm. In 1935 the present author pointed out that the cytoplasm at the cleavage planes does not contain the pigment granules (MOTOMURA '35, '36). This suggests the new formation of the cleavage plane. The possibilities of the new formation of the cleavage plane were enumerated by the present author from an embryological view-point, and some of them were supported recently by DAN and others. From the above mentioned facts the present author is inclined to believe that the new formation of the cleavage plane begins at the second period of the cleavage.

Recently SCHECHTMAN ('37) obtained very interesting results on the

mechanism of cleavage in an amphibian egg by means of the local vital staining method. According to him, the cleavage plane of this form is covered by extension of the material of the cortical cytoplasm of the furrow region. The result seems to differ from that in the case of a sea urchin, *Strongylocentrotus pulcherrimus*, which has a natural color mark in the cortical cytoplasm of the egg. This subject must be settled in the future.

In the present study, the method of kaolin marking, which was first applied by DAN, YANAGITA and SUGIYAMA, was used. The results mentioned above are only reliable when the hyaline membrane exactly follows the behavior of the cortical cytoplasm. As was discussed by DAN and others ('37, '38), the slipping between the hyaline membrane and the plasma membrane is not possible in the sea urchin egg. A decisive answer in regard to this point may be obtained by making the marks simultaneously on the hyaline and plasma membranes.

SUMMARY

Changes in surface area of different regions of the eggs of a sea urchin, *Temnopleurus hardwickii*, in the course of the first cleavage were studied. The position of kaolin marks attached to the hyaline membrane of the egg was traced every minute by camera lucida drawings. And the areas of the proximal and distal portions of the egg which were divided by the kaolin mark were measured graphically.

From the changes of the surface areas, three periods are distinguishable in the time of the first cleavage, which is from the first appearance of the first cleavage furrow till the beginning of the second cleavage furrow. In the first or constriction period, the apparent completion of the cleavage furrow and the extension of the polar region were observed. In the second period the contraction of the polar region and the increase of the area of the furrow region occur. In the final period the surface remains stationary till the beginning of the second cleavage furrow.

REFERENCES

- DAN, K., YANAGITA, T. and SUGIYAMA, M. 1937. Behavior of the Cell Surface During Cleavage. I. *Protoplasma* **28**, 66.
DAN, K., DAN, J. C. and YANAGITA, T. 1938. ditto II. *Cytologia* **8**, 521.
MOTOMURA, I. 1933. On the Presence of the Immovable Cortical Cytoplasm in the Centrifuged Sea Urchin Egg and its Importance on the Determination of the Polarity (Preliminary Report). *Sci. Rep. Tôhoku Imp. Univ. Biol.* **8**, 255.

- MOTOMURA, I. 1935. Determination of the Embryonic Axis in the Eggs of Amphibia and Echinoderms. *ibid.* **10**, 211.
- 1936. On the Cytoplasmic Structure of the Egg of a Sea Urchin *Strongylocentrotus pulcherrimus* from the View Point of Embryology. *Zool. Mag. Tokyo.* **48**, 90.
- SCHECHTMAN, A. M. 1937. Localized Cortical Growth as the Immediate Cause of Cell Division. *Science* **85**, No. 2200, 222.
- SPEK, J. 1918. Oberflächenspannungsdifferenzen als eine Ursache der Zellteilung. *Roux' Arch.* **44**, 1.

REPORT ON THE CALCAREOUS SPONGES OBTAINED
BY THE ZOOLOGICAL INSTITUTE AND
MUSEUM OF HAMBURG
PART I.

By

SANJI HÔZAWA

Biological Institute, Tôhoku Imperial University, Sendai, Japan

(With Plates VI-VII and 10 text-figures)

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The present paper deals with the calcareous sponges which were collected by the Zoological Institute and Museum of Hamburg. They were sent to the present writer to examine by the courtesy of Dr. THIEL of the same Institute and Museum.

The sponges were secured from various widely distributed localities, such as Mexico, Naples, Messina, Scilly, Haiti Island, Seychell Island in Indian Ocean, Portugal, Japan and the Southern end of South America in the neighbourhood of the Magellan Straits.

In the first part of the report the writer has treated all the specimens from the above localities except those from the Southern end of South America in the neighbourhood of the Magellan Straits. These will be reported upon in the second part of this paper in the near future.

The number of species here reported on is 14 in all, and they belong to 8 genera and 5 families.

Of these 14 species, 8 are those previously known and the remaining 6 are new to science.

The following is the list of species.

Family Homocoelidae

- 1) *Leucosolenia complicata* (MONTAGU)
- 2) *Leucosolenia canariensis* (MICHLUCHO-MACLAY)

Family Leucaltidae

- 3) *Leucaltis clathria* HAECKEL

Family Syceetidae

- 4) *Sycon mexicanum*, n. sp.
- 5) *Sycon coronatum* (ELLIS et SOLANDER)

Family Heteropiidae

- 6) *Vosmaeropsis japonica* HÔZAWA
- 7) *Vosmaeropsis simplex*, n. sp.
- 8) *Vosmaeropsis levis*, n. sp.
- 9) *Vosmaeropsis triradiata*, n. sp.
- 10) *Amphiute paulini* HANTSCH

Family Grantiidae

- 11) *Grantia mexica*, n. sp.
- 12) *Anaximilla irregularis* BURTON
- 13) *Leucandra pumila* BOWERBANK
- 14) *Leucandra seychellensis*, n. sp.

Family Homocoelidae DENDY

Genus LEUCOSOLENIA BOWERBANK

1) *Leucosolenia complicata* (MONTAGU)

(Pl. VI, Fig. 1; textfig. 1)

Spongia complicata. MONTAGU, 1812, p. 97, Pl. IX, figs. 2, 3.

Grantia botryoides, LIEBERKÜHN, 1859, p. 373.

Olynthus hispidus, HAECKEL, 1870, p. 237.

Leucosolenia complicata, MINCHIN, 1905, pp. 360-373, textfigs. 91-93; DENDY and ROW, 1913, p. 721.

This species is represented by a unique specimen (Spec. No. S. 1232) in the collection.

The sponge-colony forms an irregular mass, composed of a loose network of branching and anastomosing Ascon-tubes. Some of these branches are provided with oscula at their apex, while some others terminate quite blindly. The oscula mentioned above are naked and are circular in outline with a diameter up to 0.5 mm. The diameter of the cylindrical Ascon-tubes varies in different parts of the colony, measuring from 0.3 to 0.8 mm. The outer surface of the Ascon-tubes appears distinctly hispid to the naked eye, and the gastral surface also appears hispid under the microscope.

The colour in alcohol is white. The texture is fairly elastic.

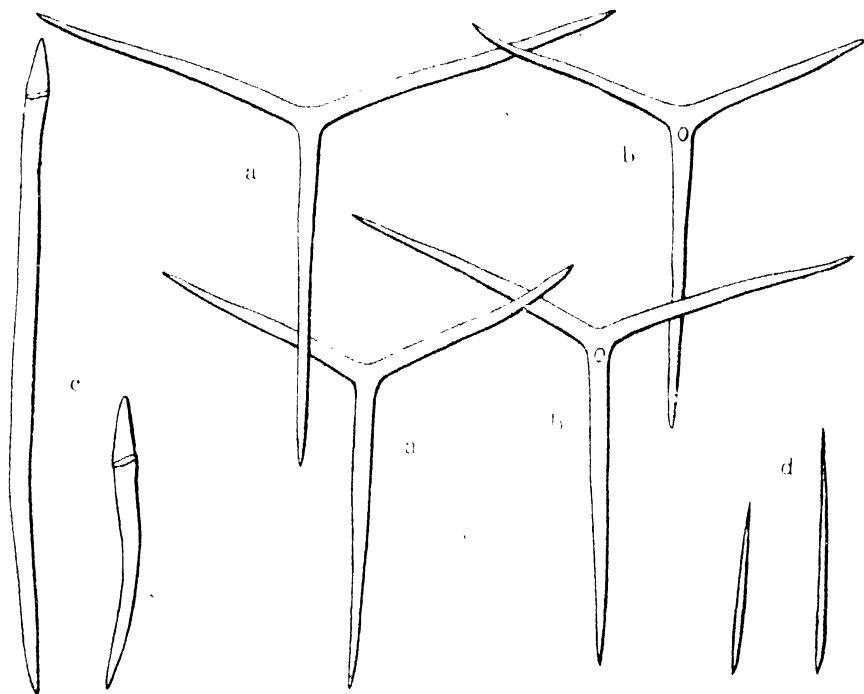
Structure: — The skeleton of the Ascon-tubes consists of triradiates, quadriradiates and oxea. The former two kinds of spicules are arranged in a thin layer, being placed close together with rays overlapping. The quadriradiates are more numerous than the triradiates and their apical rays project into gastral cavity. There exist two kinds of oxea. The

large oxea project more or less from the outer surface of the tubes at varying angles and give a hispid appearance to the latter. Their inner ends also often project into the gastral cavity. The small and slender oxea also project here and there from the outer surface of the Ascon-tubes.

Spicules (textfig. 1): — Triradiates slightly sagittal (a). All rays are equally thick and sharply pointed. Basal ray quite straight, mostly longer than the paired rays or at least equal to them in length, about 100μ long and about 8μ thick at the base. Paired rays show a double curvature, proximally they slope backwards and distally they curve forwards, $80\text{--}100\mu$ long and about 8μ thick at the base.

Quadriradiates (b) similar to the triradiates, except in the presence of apical ray. Apical ray distinctly shorter and slightly thinner than facial rays, being slightly curved upward, broadest at base and narrowing distally to terminate in a sharp point, $30\text{--}50\mu$ long and about 4μ thick at the base.

Large oxea (c) slightly curved and are of a more or less irregular outline, proximally tapering to a sharp point, distally terminating with a



Textfig. 1. *Leucosolenia complicata* (MONTAGU). a, triradiates of Ascon-tube; b, quadriradiates of the same; c, large oxea; d, small oxea (all $\times 400$).

lance-head which is provided with sharp or obtuse apex, 100–220 μ long and about 10 μ thick at the thickest part.

Small and slender oxea (d) straight, thickest close to the proximal end, where it tapers rapidly to a point. From the region of greatest thickness the shaft taper gradually to the very sharp distal extremity, 50–80 μ long and about 3 μ at the thickest part.

Localities: - Roskoff, Atlantic Coast of France (MINCHIN); Scilly.

Remarks: *Leucosolenia complicata* is a species first described by MONTAGU and recorded since by several investigators.

In 1905 MINCHIN contributed an article entitled "Characters and Synonymy of the British Species of the Genus *Leucosolenia*". In that paper he dealt also with the species now described and he fully discussed the external characters, the nature of the spiculation, etc. The writer has identified the sponge which was taken from the Scilly Islands with the present species now described. The external and internal features of the specimen seem to agree very well with the descriptions and figures made of this species by previous writers.

2) *Leucosolenia canariensis* (MICHLUCHO-MACLAY)

(Pl. VI, Fig. 2; textfig. 2)

Nardoa canariensis, MICHLUCHO-MACLAY, 1868, p. 230.

Nardoa sulphurea, MICHLUCHO-MACLAY, 1868, p. 230.

Nardoa rubra, MICHLUCHO-MACLAY, 1868, p. 230.

Torroma canariense, HAECKEL, 1870, p. 244.

Torroma rubrum, HAECKEL, 1870, p. 245.

Ascaltis canariensis, HAECKEL, 1872, p. 52, pl. 9, figs. 1–3; pl. 10, figs. 1, a c.

Ascaltis compacta, SCHUEFFNER, 1877, p. 404, pl. 25, fig. 9.

Leucosolenia nanseni, BREITFLUSS, 1896, p. 427; 1898, p. 106, pl. 12, figs. 1–9.

Leucosolenia canariensis, LAKSCHWITSCH, 1886, p. 300, pl. 7, fig. 1; THACKER, 1908, p. 762, pl. 40, fig. 3, textfigs. 157–160; DENDY and ROW, 1913, p. 724; HÔZAWA, 1918, p. 528; 1933, p. 2. Pl. I. fig. 1.

The collection contains seven specimens of this species, all growing attached to the same foreign body.

They differ considerably from each other in size ranging from 2 mm. to 15 mm. in diameter. They differ also in shape varying from a more or less oval body to lobular masses of irregular shape. From the lower surface of the sponge arise a few short root-like processes which serve to attach the sponge body to the foreign body.

The sponge consists of a network of branching and anastomosing Ascontubes leaving pseudopores on the outer surface. The pseudopores vary

in size up to about 0.6 mm. in diameter and are mostly of oval or elongate oval shape. The Ascon-tubes average 0.2-0.3 mm. in diameter. No oscula could be distinguished on the sponge surface.

The colour of the sponge is brownish white. In texture it is soft, but fairly tough and not very easily torn.

Skeleton: -- The skeleton consists of triradiates and quadriradiates. The triradiates are arranged in an irregular and almost in a single layer in the walls of the Ascon-tubes. The quadriradiates are relatively few but easily discoverable. They occur here and there among the triradiates above mentioned with their apical rays projecting into the gastral cavity.

Spicules (textfig. 2): -- Triradiates (a) regular. Rays conical gradually sharp-pointed, measuring about 80μ in length and about 6μ in thickness.

Quadriradiates (b) about the same size and shape as triradiates, but with an apical ray projecting at right angles from the centre. It is quite straight and is nearly as long and thick as facial rays.

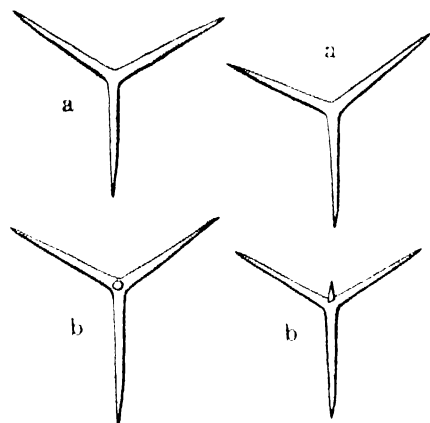
Previously known Distribution: -- Canary Islands (MICHLUCHO-MACLAY); Cape Verde Islands (THACKER); Mauritius (SCHNUFFNER); Minorca (LACKSCHEWITSCH); Spitzbergen, Arctic Ocean (BREITFUSS); Off the north point of Copper, Island, Commander Islands; Off Yuriage, Rikuzen, Japan (HÔZAWA).

Locality: -- Mexico.

Remarks: As the writer stated in a previous report (1933) the present species seems to be widely distributed all over the world, and may thus be considered to be cosmopolitan.

In 1908 THACKER gave a full account discussing the morphological features, the synonymy and the affinities of this species. In that account he has mentioned that there is not much variation in the size of the spicules in the same individual, but much difference between the spicules of different specimens. The writer was also

able to notice the same fact in the cases of the three specimens from Copper Island, Japan and Mexico respectively.



Textfig. 2. *Leucosolenia canariensis* (MICHLUCHO-MACLAY). a, triradiates of Ascon-tube; b, quadriradiates of the same (all $\times 200$).

The first two specimens have already been dealt with by the writer in his previous papers. In the present specimen from Mexico the spicules are much smaller and slender than those of the first two and are distributed more sparsely.

Family Leucaltidae DENDY and ROW

Genus LEUCALTIS HAECKEL

3) *Leucaltis clathria* HAECKEL

(Pl. VI, Fig. 3)

Leucaltis clathria, HAECKEL, 1872, p. 159-161, Taf. 28, figs. 3 a-3 c; DENDY, 1913, p. 16, pl. 2, figs. 1, 2; DENDY and ROW, 1913, p. 738.

Heteropegma nodus gordii, POLÉJAEFF, 1883, pp. 45-46 pl. 1, fig. 7, pl. IV, figs. 1 a-1 d; VON LENDENFELD, 1885, p. 1107-1109; DENDY, 1892, p. 113; 1893, p. 204, pl. 13, fig. 20; 1905, p. 230; HANITSCH, 1895, p. 209; JENKIN, 1908, p. 153, textfig. 103.

Leucaltis bathybia var. *mascarenica*, RIDLEY, 1884.

Clathrina latitubulata, CARTER, 1886, p. 515-516.

Heteropegma latitubulata, DENDY, 1892, p. 114.

In the collection there are several specimens of this species contained in the same bottle. They are unfortunately broken into pieces and thus it is impossible to know their original external shape. But judging from the fact that in the bottle some Madreporarian Coral is found to which some sponge fragments are attached, it may be easily recognizable that the sponge had grown attached to these corals and that it had consisted of an irregular mass of branching and anastomosing tubes.

In most cases the tubes are strongly laterally compressed and are provided with many small circular oscula distributed at intervals. The diameter of the tubes measures from 2 mm. to 8 mm. and the thickness of the wall is about 0.5 mm.

With respect to the minor structure it has been fully recorded by previous investigators such as HAECKEL, POLÉJAEFF, JENKIN, DENDY, etc. and therefore it may be conceded that no further details are needed here.

Previously known Distribution:—Coast of Florida (HAECKEL); Off Bermudas (POLÉJAEFF); Cape York, Torres Straits (POLÉJAEFF); near Port Phillip Heads (CARTER, DENDY); Ceylon (DENDY); West Coast of Portugal (HANITSCH); Amirante Group, Seychelles (RIDLEY, DENDY); Wasin E. Africa (JENKIN); Cargodas Carajos (DENDY); Egmont Reef (DENDY).

Locality:—Seychelles Islands, Indian Ocean.

Remarks:—This species was first described by HAECKEL in 1872,

the specimen being obtained on the coast of Florida.

In 1913, DENDY identified his specimens from Cargodos Carajos, Amirantes, and the Egmont Reef with this species.

In the same report he has discussed that *Heteropegma nodus gordii* which was first named by POLÉJAEFF in 1883 and afterwards recorded by LENDENFELD (1885), DENDY (1892, 1893, 1905), HANITSCH (1895), JENKIN (1908) is synonymous with the present species.

That both of *Leucaltis bathybia* var. *mascarenica* RIDLEY and *Clathrina latilubulata* CARTER are synonymous is also mentioned in his same report.

RIDLEY reported this species as having been found in the Seychelle Islands in the Indian Ocean and this is the second case of the species being recorded as having been found in that locality.

Family Sycettidae DENDY

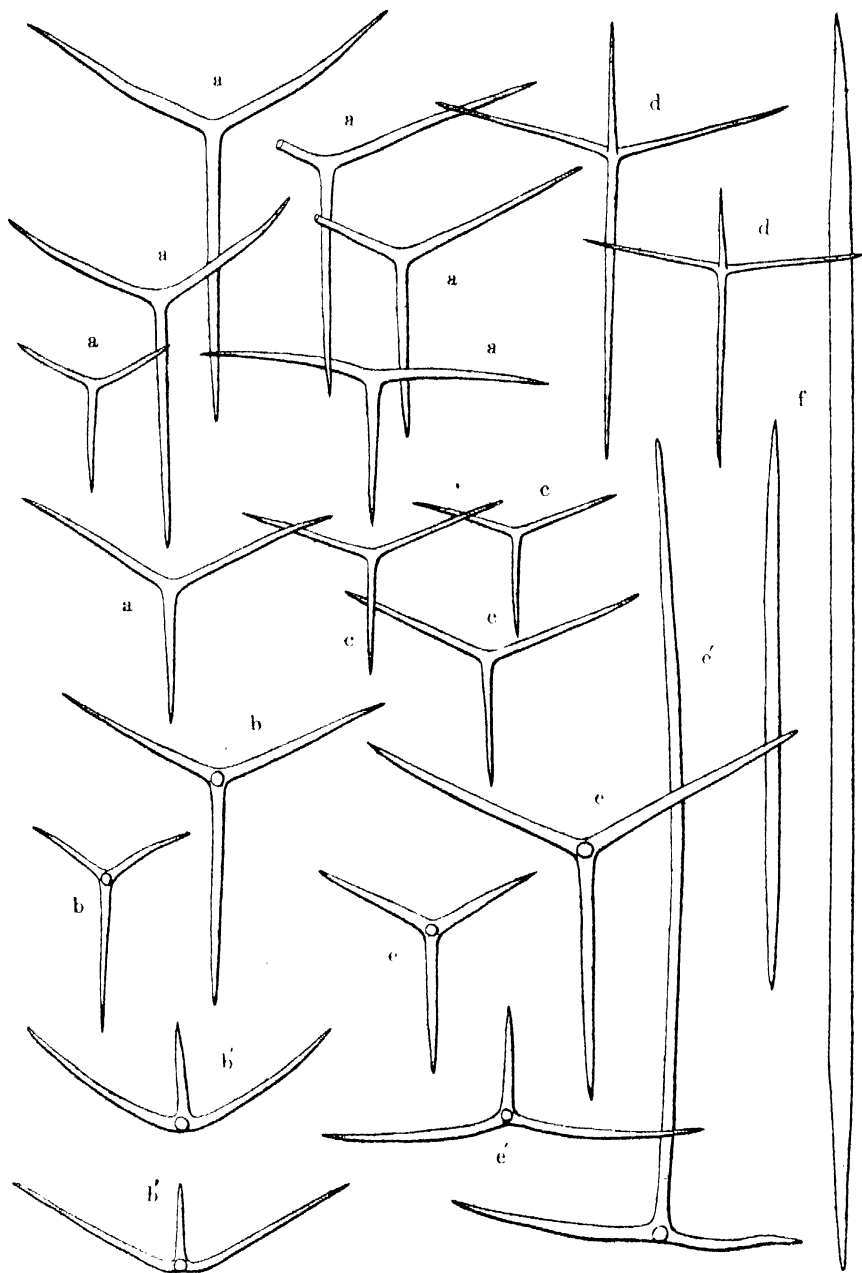
Genus SYCON RISSO

1) *Sycon mexicanum*, n. sp.

(Pl. VI, Fig. 4; textfig. 3)

There is a single specimen (Spec. No. 1752) of this new species in the collection. It has the form of an elongate sac with an oval osculum at the upper end which is surrounded by a well-developed collar. The total length of the body is 12 mm. including the oscular collar of about 1.2 mm. high and the breadth is about 5 mm. The sponge wall is about 1 mm. thick in the lower parts of the body and becomes gradually thinner nearer the osculum. The outer surface of the sponge is of a decided hairy appearance on account of the presence of projecting tufts of long oxea at the distal ends of flagellated chambers. The gastral surface, when observed under the microscope, is strongly hispid from the long apical rays of gastral quadriradiates. The colour in alcohol is greyish white and the texture is rather soft.

Structure: - The canal system is typical. The flagellated chambers are straight and not branching. They touch each other with all of their length and have small projecting distal ends crowned by tufts of long oxea. The interspaces distributed among the distal ends of flagellated chambers looks to form a thin dermal cortex showing the transition from the Sycettidae to the Grantiidae. The tubar skeleton is composed of the basal rays of subgastral quadriradiates and of triradiates arranged in several layers. A few of these radiates appear to be furnished with the apical ray projecting into the flagellated chamber. The basal rays of the



Textfig. 3. *Sycon mexico*, n. sp. a, tubar triradiates; b, tubar triradiates, b', the same seen from above; c, triradiates forming dermal skeleton; d, subgastral quadriradiates; e, gastral quadriradiates; e', the same seen from above; f, large oxea (all $\times 150$).

outermost triradiates which project with the oxea and form the distal tufts. The gastral skeleton is formed of the paired rays of subgastral quadriradiates and of gastral quadriradiates which are tangentially placed with their long apical rays projecting into the gastral cavity. The apical rays of subgastral quadriradiates are turned towards the exhalant canal, so that when looked from the gastral surface they appear to form a protection to the apophyle. The skeleton of oscular collar is a close interlacement of triradiates and quadriradiates, both of which have strongly divergent paired rays and downwardly directed basal ray. There exist in addition some oxea, the slender and the thicker, in longitudinal disposition.

Spicules (textfig. 3): — Tubar triradiates (a) with slender rays of nearly equal thickness, not lying in the same plane. The oral angles are variable, being somewhat wider in the spicules found in the inner parts of the sponge wall than those in the periphery. Basal ray straight not strongly differentiated in length from paired rays, but slightly thinner than the latter, 100–235 μ long and 8–10 μ thick at the base. Paired rays equal or subequal, rather widely divergent, 78–180 μ long and 8–10 μ thick at the base.

Tubar puadriradiates (b) almost similar to the tubar triradiates but with an apical ray which is distinctly shorter than facial rays. Apical ray is 62–94 μ long and 8–10 μ thick at the base.

Triradiates forming the dermal skeleton (c) smaller and thinner than tubar triradiates. Rays nearly straight not strongly differentiated and are of equal thickness. Basal ray, 78–112 μ long and 6–8 μ thick at the base. Paired rays widely divergent, 90–123 μ long and 6–8 μ thick at the base.

Subgastral quadriradiates (d) sagittal, slender-rayed. Paired rays and apical ray lie in one plane and the basal ray stands at right angle to that plane. Basal ray longer and slightly thinner than paired rays, straight in facial view, but slightly curved at base when observed laterally, 168–224 μ long and about 6 μ thick at the base. Paired rays slightly longer than apical ray, very widely divergent, 125–156 μ long and 8 μ thick at the base. Apical ray sharply pointed 78–95 μ long and about 6 μ thick at the base.

Gastral quadriradiates (e) sagittal. Rays straight in facial view. Basal ray slightly longer than paired rays, 112–200 μ long and 10–12 μ thick at the base. Paired rays 95–63 μ long and 10–12 μ thick at the base.

Large oxea at the distal end of flagellated chamber (f) nearly straight and uniformly thick in the greater part of their length but tapering at both sharply pointed ends, 480 μ –1.1 mm long and 10–18 μ thick in the

middle.

Linear spicules at the distal end of flagellated chamber sharply pointed at both ends and are variable in both length and thickness.

Triradiates of oscular collar strongly sagittal.

Quadriradiates of oscular collar nearly same as the triradiates of the same except for the presence of apical ray.

Locality: — Mexico.

Remarks: — This species seems to be quite distinct from any of the hitherto known species.

It is remarkable for the presence of subgastral quadriradiates as well as of a feebly developed dermal cortex which is provided with its proper skeleton. Other members of the genus such as *Sycon stauriferum* (PREIWITSCH)¹, *Sycon australe* (JENKIN)², *Sycon setosum* O. SCHMIDT³, *Sycon yatsui* HÔZAWA⁴, *Sycon digitiformis* HÔZAWA⁵ have also the subgastral quadriradiates, but they have no dermal cortex.

5) *Sycon coronatum* (ELLIS and SOLANDER)

(Pl. VI, Fig. 5; textfig. 4)

Spongia coronata, ELLIS and SOLANDER, 1786, p. 190, Tab. 58, figs. 8, 9.

Sycandra coronata, HAECKEL, 1872, p. 301, Taf. 51, fig. 2 a-t; Taf. 60, figs. 1-6.

Syncon coronatum, DENDY, 1892, p. 79; DENDY and ROW, 1913, p. 745; BREITFUSS, 1935, pp. 16-17.

In the collection there are ninety one specimens of this sponge, all of which being obtained from Messina and are preserved in one bottle numbered S. 1080.

They are variable in shape ranging from sac-like to elongate tube-like and are variable in size as well. They are 2.5-12.8 mm. in total length including the oscular fringe of 0.9-2.5 mm. high, and are 1.1-6.6 mm. in the greatest breadth.

The largest specimen measuring 12.8 mm. in total length and 6.6 mm. in the greatest breadth was herewith chosen as one on which to base further description.

It forms a solitary tubular individual broadest near the base, and is

¹ *Sycandra stauriferu* PREIWITSCH, 1904 pp. 17-19, Taff. III, fig. 8.

² *Streptoconus australis* JENKIN, 1908 pp. 25-27, pl. XXVII, fig. 3; pl. XXXII, XXXIII, figs. 75-80.

³ *Sycon setosum* O. SCHMIDT, 1862 p. 15, Taf I, fig. 3,

⁴ *Sycon yatsui* HÔZAWA, 1929 pp. 297-300, Fig. 14, 15; textfig. 8.

⁵ *Sycon digitiformis* HÔZAWA, 1929 pp. 307-310, figs. 22, 23; textfig. 12.

provided with an osculum which is surrounded by a collar at the upper end. The oscular collar is about 2.5 mm. high and is 1.5 mm. across. The wall is about 2 mm. thick in the basal parts and gradually diminishes in thickness towards the thin oscular margin.

The dermal surface appears strongly hairy on account of the projecting tufts of long oxea at the distal ends of flagellated chambers. The gastral surface appears nearly smooth to the naked eye, but it is minutely punctate due to the angular openings of exhalant canals and appears hispid under the microscope from the projecting apical rays of gastral quadriradiates.

Colour in alcohol is grevish white and the texture is fairly rigid and elastic.

Structure : -- Canal system is typical. The flagellated chambers straight, nearly equally thick throughout their entire length terminating in low rounded distal cones.

The tubar skeleton is composed of the basal rays of subgastral triradiates and quadriradiates as well as several layers of sagittal triradiates arranged in a typical manner. Some of these triradiates are provided with a short apical ray projecting into the flagellated chamber. At the distal end of flagellated chambers there occur two kinds of oxea, the thicker and the thinner, forming thick tufts.

The gastral skeleton is made up of quadriradiates arranged tangentially in a few layers and with their basal rays projecting towards the sponge base. To the skeleton the facial rays of subgastral tri- and quadriradiates are to be added. The apical rays of subgastral quadriradiates project into the exhalant canal.

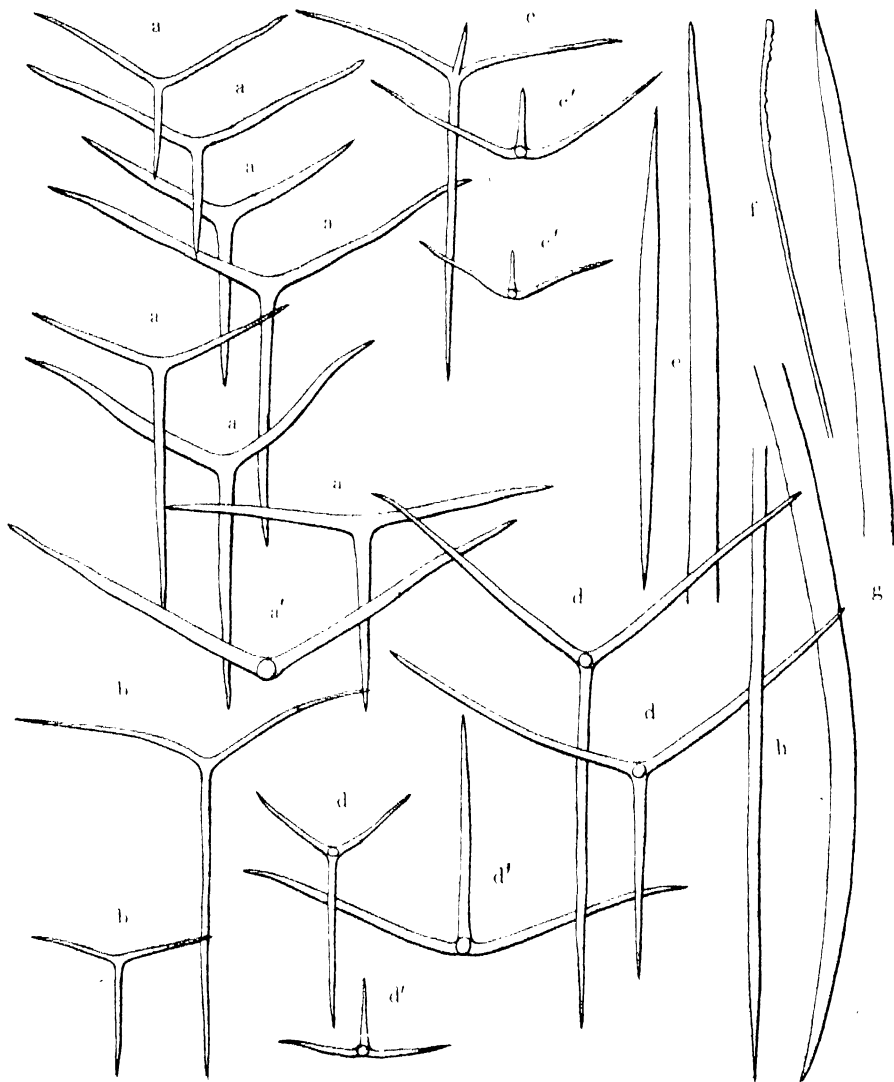
The skeleton of the oscular collar is composed of quadriradiates and oxea. The first kind of spicule is different from these found in the gastral skeleton in having strongly divergent paired rays and in having been set closely.

Spicules (textfig. 4) : -- Tubar triradiates (a) slightly sagittal, rather variable in size and shape. Rays are of nearly equal thickness and not lying in the same plane. The oral angles are variable being wider in the spicules found in the inner parts of the sponge wall than in those in the periphery. Basal ray usually longer and sometimes shorter than paired rays, straight, 180–290 μ long and 6–16 μ thick at the base. Paired rays more or less widely diverging, slightly curved forwards, 140–240 μ long and 6–16 μ thick at the base.

Subgastral triradiates (b) strongly sagittal. Rays nearly equally thick. Basal ray straight, slightly longer than paired rays, 80–250 μ long and

6–8 μ thick at the base. Paired rays very widely diverging, slightly curved backwards, 65–150 μ long and 6–8 μ thick at the base.

Subgastral quadriradiates (c) almost like subgastral triradiates with



Textfig. 4. *Sycon coronarum* (ELLIS and SOLANDER). a, tubar triradiates; a', the same seen from above; b, subgastral triradiates; c, subgastral quadriradiate; c', the same seen from above; d, gastral quadriradiates; d', the same seen from above; e, oxea at distal end of flagellated chamber; f, hair-like oxea at the same; g, oxea of oscular margin; h, hair-like oxea forming the oscular collar (all $\times 150$).

addition of an apical ray which is shorter than either the paired rays or the basal ray. Apical ray straight $35-55\ \mu$ long and $6-8\ \mu$ thick at the base.

Gastral quadriradiates sagittal (d). Basal ray straight, generally longer than paired rays, but often shorter, $150-310\ \mu$ long and $6-8\ \mu$ thick at the base. Paired rays nearly equal, slightly curved forwards in basal parts and either straight or very slightly flexing backwards in the remaining parts, $70-210\ \mu$ long and $6-8\ \mu$ thick at the base. Apical ray well-developed, not strongly differentiated in length and thickness from facial rays, $62-210\ \mu$ long and $6-8\ \mu$ thick at the base.

Oxea at distal end of flagellated chamber (e) generally slightly curved, uniformly thick throughout their greater length, sharply pointed at both ends, $400\ \mu-1.5\ \text{mm.}$ long and $14-22\ \mu$ thick.

Hair-like oxea at distal end of flagellated chamber (f) very slender, straight or slightly curved, nearly uniformly thick in the middle greater parts and sharply pointed at both ends, $260\ \mu-1.5\ \text{mm.}$ long and about $2\ \mu$ thick.

Oxea of oscular margin (g) almost similar to those found at distal end of the flagellated chambers but are straighter, $500\ \mu-1.5\ \text{mm.}$ long and $14-26\ \mu$ broad.

Hair-like oxea forming the oscular collar (h), rather slender, straight, nearly uniformly thick in the greater part of their length though tapering at ends which are fairly sharply pointed, $700\ \mu-4\ \text{mm.}$ long and $2-8\ \mu$ thick.

Previously known Distribution: East coast of Australia; Mediterranean; Atlantic Ocean; Pacific Ocean; Indian Ocean.

Locality: -- Messina.

Family Heteropiidae DENDY

Genus VOSMAEROPSIS VON LENDENFELD

6) *Vosmaeropsis japonica* HÔZAWA

(Pl. VI, Fig. 6)

Vosmaeropsis japonica HÔZAWA, 1929, p. 324, figs 34, 35, textfig. 18.

Of this species there exists in the collection only a single specimen (Spec. No. 1236) which was obtained from the Sagami, Sea, Japan.

It is in the form of an irregularly curved tube narrowed towards the attachment base, and showing near the upper end an oval osculum of about $1\ \text{mm.}$ diameter.

The specimen is about 13 mm. in length and about 8 mm. broad in the broadest part.

This species was first described by the writer in 1929, the type specimen being taken from Misaki, on the coast of the Sagami Sea. Thus the present report notes the occurrence of this species in the Sagami Sea for the second time.

With respect to the structure, spicules, etc. the present specimen is almost similar to the type specimen.

Locality: — Sagami Sea, Japan.

7) *Vosmaeropsis simplex*, n. sp.

(Pl. VI, Fig. 7; textfig. 5)

In the collection there exists only a single specimen (Spec. No. S. 215) of this new species, and it was obtained at Naples.

The sponge is in form an irregularly shaped mass consisting of a small number of compressed tubes. The length and the breadth of the mass are respectively 60 mm. and 40 mm. The thickness of the wall of the tube is thickest in the basal part measuring about 5 mm. but it becomes thinner towards the oscular margin. The dermal surface of the sponge appears nearly smooth to the naked eye though it is not entirely even. The gastral surface is also smooth but is perforated by many exhalant apertures irregularly distributed.

The colour is greyish white in alcohol and the texture is fairly rigid and is rather harsh to touch.

Structure: The canal system of this species is of the leuconoid type. The flagellated chambers are of sac-like shape, measuring about 120μ in diameter. The dermal skeleton is composed of triradiates of two kinds, the larger and the smaller, both of which are tangentially but confusedly arranged in a small number of layers. There may be added the paired rays of pseudosagittal triradiates.

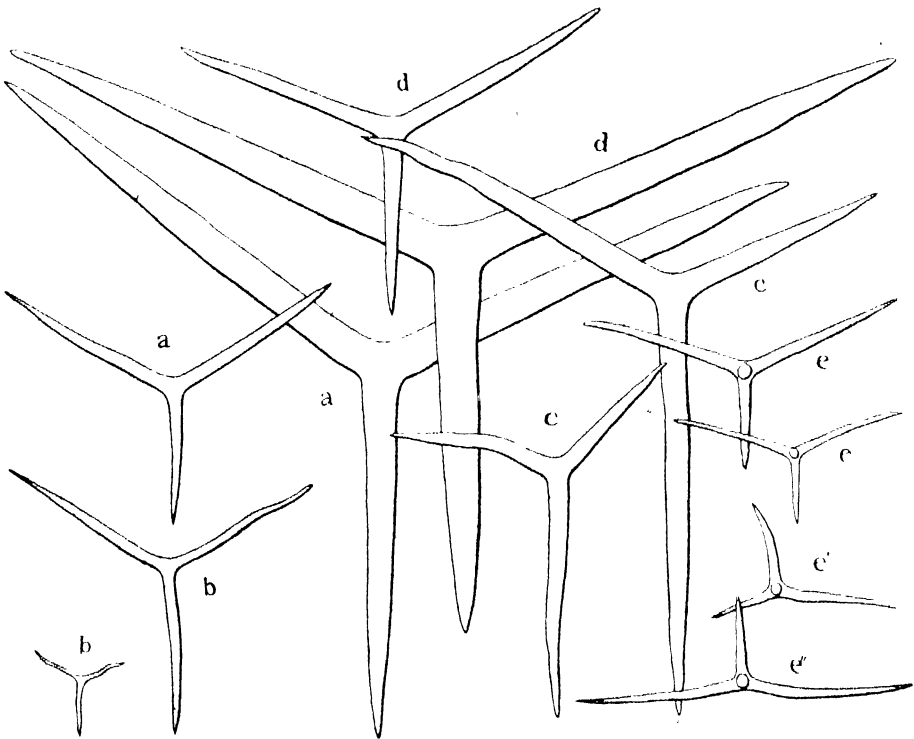
The tubar skeleton is composed of the basal rays of subdermal pseudosagittal triradiates and of the triradiates irregularly disposed. Some of these spicules take the subdermal position with their basal rays pointing centrifugally. The gastral skeleton is made up of quadriradiates arranged in a few layers. They are placed rather irregularly and their apical rays project into the gastral cavity. A small number of triradiates in a scattered distribution may be added to the same skeleton.

Spicules (textfig. 5): — Larger dermal triradiates (a) slightly sagittal

with rays of equal thickness. Basal ray straight, slightly shorter than paired rays, being $160\text{--}470\ \mu$ long and $20\text{--}45\ \mu$ thick at the base. Paired rays are sometimes slightly differentiated in length, being $230\text{--}600\ \mu$ long and $20\text{--}45\ \mu$ thick at the base.

Smaller dermal triradiates (b) slightly sagittal with rays of equal thickness and disposed slightly convexly towards the outer side. Basal ray nearly straight or slightly curved, being $54\text{--}200\ \mu$ long and $8\text{--}16\ \mu$ thick at the base. Paired rays slightly curved forwards in greater part of the basal and either straight or irregularly curved in the distal part, being $74\text{--}212\ \mu$ long and $8\text{--}16\ \mu$ thick at the base.

Subdermal triradiates (c) pseudosagittal. All rays equally thick, lying nearly in the same plane and are slightly wavy in outline. Basal ray nearly straight, distinctly longer than paired rays being $320\text{--}530\ \mu$ long and $28\text{--}38\ \mu$ thick at the base. Paired rays are unequal in length and



Textfig. 5. *Vasmaeropsis simplex*, n. sp. a, larger dermal triradiate; b, smaller dermal triradiates; c, subdermal triradiates; d, tubar triradiates; e, gastral quadriradiates, e', the same seen from side; e'', the same seen from above (all $\times 100$).

shape. The longer ray usually curved, gradually tapering, $190-425\ \mu$ long and $26-32\ \mu$ thick at the base. The shorter ray less curved than the longer, $180-270\ \mu$ long and $26-32\ \mu$ thick at the base.

Tubar triradiates (d) slightly sagittal, all rays being equal in thickness. Basal ray straight, shorter than paired rays, $230-470\ \mu$ long and $24-56\ \mu$ thick at the base. Paired rays are of equal or slightly differentiated length, gradually sharply pointed, $290-620\ \mu$ long and $24-56\ \mu$ thick at the base.

Gastral quadriradiates (e) sagittal. All rays equally thick. Basal ray much shorter than paired rays, quite straight, tapering from base to the sharp point, $75-100\ \mu$ long and $10-16\ \mu$ thick at the base. Paired rays subequal in length, slightly curved backwards, $126-210\ \mu$ long and $10-16\ \mu$ thick at the base. Apical ray curved, sharply pointed, much shorter and slightly thinner than paired rays, $97-110\ \mu$ long and $10-14\ \mu$ thick at the base.

Locality: Naples.

Remarks:—This new species may be easily distinguished from most members of the genus *Vosmaeropsis* in having no oxea of any kind.

*Vosmaeropsis maculata*¹⁾ obtained from Japan also has not oxea of any kind but it is distinguishable from the species now being described by the spicular arrangement, by the characteristics of the spicules and by the external form.

The specific name of this new species was given on account of the fact that it is rather simple in structure compared with other members of the same genus.

8) *Vosmaeropsis levis*, n. sp.

(Pl. VII, Fig. 8; textfig. 6)

This new species is represented by a single specimen (Spec. No. 175), which was obtained from Mexico.

It is a single individual of a somewhat curved and laterally compressed, elongated cylindrical form, broadest at the part a little below the middle. The total length is about 18 mm. and the greatest breadth is about 8 mm. The osculum at the upper end is oval with a longer diameter of 1.5 mm. and shows neither an oscular fringe nor a distinct collar. The gastral cavity is deep extending throughout the greater length of the sponge. The lower part of the sponge is narrowed in a stalk-like manner.

¹⁾*Vosmaeropsis maculata*, HÔZAWA, 1929, pp. 321-324, figs. 32, 33; textfig. 17.

The dermal surface of the sponge appears on the whole smooth being deprived of projecting spicules. The gastral surface is in a punctate appearance from the exhalant apertures of various sizes, and also looks more or less hispid due to the apical rays of gastral quadriradiates. Thickness of the wall, as measured in the broadest part of the sponge body, is about 2 mm.

The colour in alcohol is greyish white and the texture is rather rigid and compact.

The canal system is of the intermediate between the syllebid and leuconid types. The exhalant canals are wide and extend through the greater part of the wall thickness. The flagellated chambers are arranged mostly around the exhalant canals and are from oval to elongate sac-like in form with a diameter of about 100-300 μ . Of these chambers those situated close to the dermal surface are more elongated than the others.

The skeleton of the dermal cortex consists chiefly of a few confused layers of triradiates placed tangentially and there may be added the paired rays of subdermal pseudosagittal triradiates. The skeleton of the chamber layer is composed of the basal rays of subdermal pseudosagittal triradiates, large triradiates in an irregular arrangement and the basal rays of subgastral triradiates. Along the larger exhalant canals there occur some quadriradiates with their apical rays projecting into the canal.

The gastral skeleton is made up of a thin layer containing the paired rays of subgastral quadriradiates as well as of gastral quadriradiates. The latter kind of spicules are rather irregularly disposed but their apical rays project into the gastral cavity.

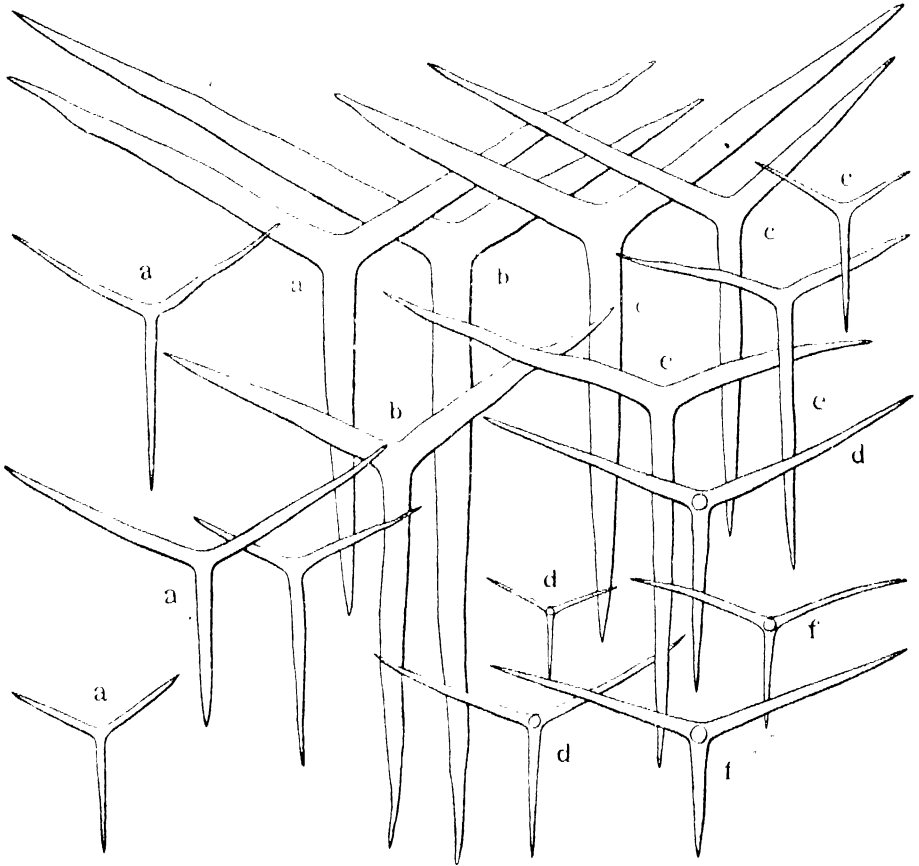
Spicules (textfig. 6) :— Dermal triradiates (a) very slightly sagittal and varies rather greatly in size. All rays nearly equally thick and gradually sharply pointed. Basal ray straight, nearly as long as or sometimes a little shorter than the paired rays, 60-450 μ long and 6-40 μ thick at the base. Paired rays slightly curved forwards, 60-470 μ long and 6-40 μ thick at the base.

Subdermal triradiates (b) pseudosagittal. All rays nearly equally thick, gradually tapering to a sharp point. Basal ray longer than paired rays, nearly straight, 240-800 μ long and 12-50 μ thick at the base. The longer ray of the paired rays nearly straight, 170-650 μ long and 12-50 μ thick at the base. The shorter of the same also nearly straight, 150-300 μ long and 12-50 μ thick at the base.

Triradiates of chamber layer (c) subregular or slightly sagittal. All rays nearly equally thick and gradually and sharply pointed. Basal ray

390–550 μ long, and paired rays 270–440 μ long, all being 30–44 μ broad at the base.

Quadriradiates of the larger exhalant canals (d) sagittal with sharply pointed facial rays of nearly equal thickness. Basal ray shorter than



Textfig. 6. *Vosmaeropsis levis*, n. sp. a, dermal triradiates; b, subdermal triradiates; c, triradiates of chamber layer; d, quadriradiates of the larger exhalant canal; e, subgastral triradiates; f, gastral quadriradiates (all $\times 100$).

paired rays, nearly straight, 90–170 μ long and 7–16 μ thick at the base. Paired rays gently curved forwards, 90–215 μ long and 6–14 μ thick at the base. Apical ray shorter and slightly thinner than facial rays, slightly curved in distal portion and is sharply pointed, 50–130 μ long and 6–14 μ broad at the base.

Subgastral triradiates (e) sagittal with rays of nearly equal thickness.

Basal ray longer than paired rays, nearly straight, about 135-490 μ long and 10-30 μ broad at the base. Paired rays strongly diverging and usually recurved, first backwards and then forwards, about 90-340 μ long and 10-39 thick at the base.

Gastral quadriradiates (f) sagittal. Basal ray distinctly shorter than paired rays and is nearly straight, 130-230 μ long and 10-20 μ broad at the base. Paired rays widely diverging, nearly straight, gradually tapering to sharp point, 185-280 μ long and 10-20 μ thick at the base. Apical ray shorter and slightly thinner than facial rays, slightly curved and sharply pointed, 80-185 μ long and 8-16 μ thick at the base.

Locality: Mexico.

Remarks: - This new species resembles *Vosmaeropsis simplex*, n. sp. which is described also in this report, in having neither oxea nor microxea. But it may be distinguished from *Vosmaeropsis simplex* by the external form, by the nature of the canal system and by the characteristics of the spicules.

9) ***Vosmaeropsis triradiata***, n. sp.

(Pl. VII, Fig. 9, textfig. 7)

Of this new species only a single specimen (Spec. No. 1775) which came from Mexico was examined by the writer. The sponge is in the form of a strongly laterally compressed oval sac of about 8 mm. high and 10 mm. broad.

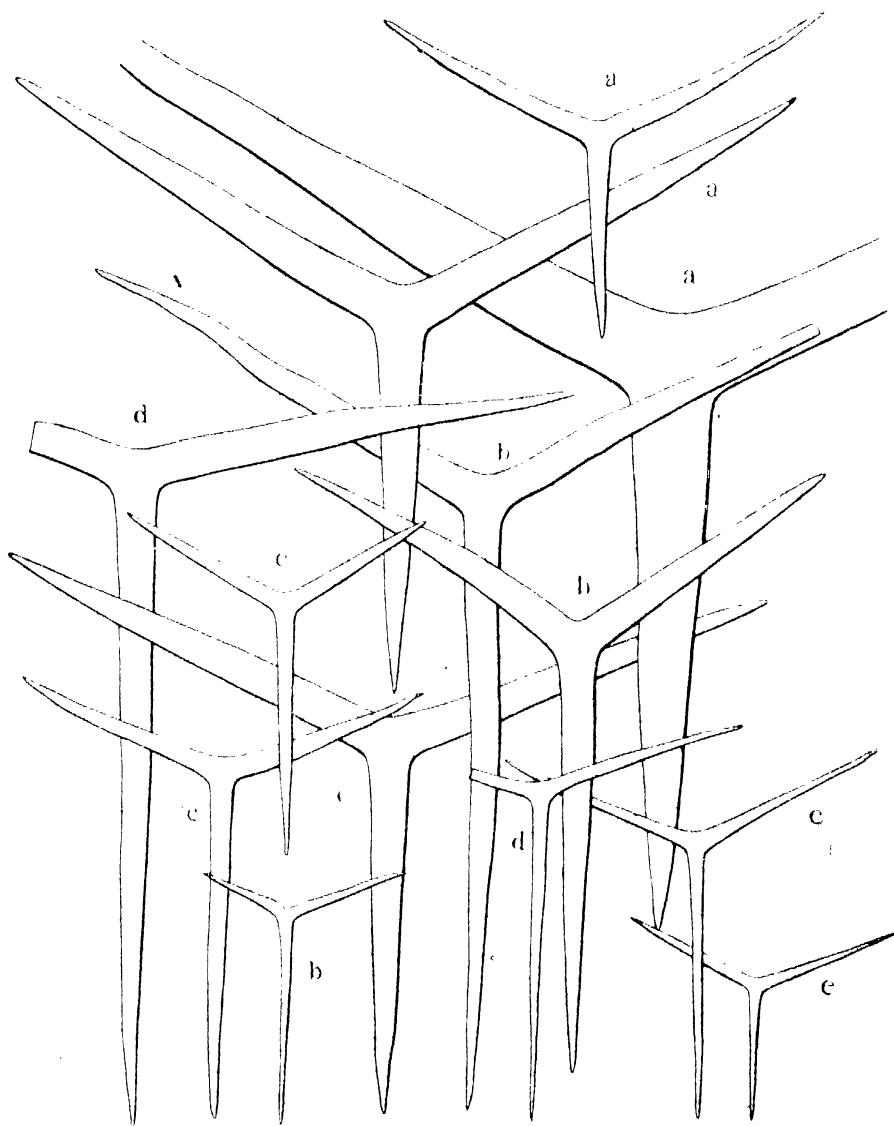
The terminal osculum leads into the gastral cavity of a habitus corresponding to that of the entire specimen. The sponge wall is thickest in the base of the body measuring about 1.5 mm. and becomes thinner towards the oscular margin. The dermal surface is also smooth though it is punctated by a great number of minute exhalant apertures uniformly distributed in mesh-like manner.

The colour in alcohol is greyish white and the texture is fairly firm and elastic.

Structure: - The canal system is of the sylleibid type though is not very typical. The flagellated chambers are variable in form ranging from an oval shape to an elongate sac-like configuration. They are arranged radially around the large exhalant canals.

The dermal skeleton consists of a dense feltwork of huge triradiate spicules arranged irregularly but parallel to the dermal surface. Besides these triradiates there may be added the paired rays of subdermal pseudo-sagittal triradiates. The skeleton of the chamber layer is made up of

the oppositely directed basal rays of subdermal and subgastral triradiates, and of some number of triradiates disposed at various levels in the chamber layer with their basal rays running centrifugally.



Textfig. 7. *Vosmaeropsis triradiata*, n. sp. a, dermal triradiates; b, subdermal triradiates; c, triradiates of chamber layer; d, subgastral triradiates; e, gastral triradiates (all $\times 100$).

The gastral skeleton is more weakly developed than the dermal. It is made up chiefly of slender triradiates arranged tangentially in a few layers and of the strongly developed paired rays of subgastral triradiates.

Around the osculum there exists not any special skeleton to be mentioned.

Spicules (textfig. 7): — Dermal triradiates (a) very large, slightly sagittal. All rays nearly equally thick and gradually sharp pointed. Basal ray straight, usually slightly shorter than paired rays, 230–760 μ long and 30–100 μ thick at the base. Paired rays slightly curved forwards, 310–980 μ long and 30–100 μ thick at the base.

Subdermal triradiates (b) pseudosagittal. All rays nearly equally thick, gradually tapering to a sharp point. Basal ray longer than the paired rays, nearly straight, 250–500 μ long and 18–40 μ thick at the base. Paired rays not strongly differentiated in length. The longer of the paired rays is 140–390 μ long while the shorter is 100–350 μ long, both being 18–40 μ thick at the base.

Triradiates of chamber layer (c) sagittal or subregular. All rays nearly equally thick and gradually tapering to sharp point. Basal ray straight, more or less longer than paired rays, 390–700 μ long and 28–50 μ thick at the base. Paired rays widely divergent, slightly curved, 265–550 μ long and 28–50 μ thick at the base.

Subgastral triradiates (d) sagittal, with sharply pointed rays of nearly equal thickness. Basal ray distinctly longer than paired rays, nearly straight, 400–780 μ long and 18–50 μ broad at the base. Paired rays strongly divergent and are slightly bent in the middle part, 250–560 μ long and 18–50 μ broad at the base.

Gastral triradiates (e) slightly sagittal with rays rather slender. Basal ray straight, slightly shorter or longer and slightly thinner than paired rays, 150–320 μ long and 12–18 μ thick at the base. Paired rays slightly curved forwards, 180–250 μ long and 14–20 μ thick at the base.

Locality: — Mexico.

Remarks: — In having neither oxea nor microxea this species resembles such species as *Vosmaeropsis samakii* HÔZAWA, *V. maculata* HÔZAWA, *V. levis*, n. sp., *V. simplex*, n. sp., but it may be easily distinguished from the said four species on account of the dermal skeleton being composed of huge triradiates and by the gastral skeleton being made up of triradiate spicules only. The specific name *triradiata* was given on account of the fact that it possesses only triradiates.

Genus AMPHIUTE HANITSCH

10) *Amphiute paulini* HANITSCH

(Pl. VII, Fig. 10)

Amphiute paulini, HANITSCH, 1895, pp. 208-209, Pl. XII, figs. 1-5, Pl. XIII, fig. 1

Only a single specimen of this species exists in the collection (Spec. No. 1181). It is a colony of two individuals, united together at their bases.

The individuals have a somewhat curved elongated cylindrical form. They are more or less laterally compressed and are narrow at their bases, but becoming gradually broader distally. In both individuals the distal parts are broken off and thus the writer is unable to discuss about the osculum. They are nearly equal in length measuring about 27 mm. while in breadth one is superior to the other, measured 4 mm. and 3 mm. respectively.

In external appearance and in the anatomical structures the present specimen represents nearly the same features as shown in the description and figures made by HANITSCH of the type-specimen.

Previously known Distribution: West coast of Portugal.

Locality: — Portugal.

Remarks: — *Amphiute paulini* is the type species of the genus *Amphiute* which was erected by HANITSCH in 1895. The genus is characterized by the possession of colossal longitudinal oxea in both dermal and gastral cortices, in addition to the presence of subdermal pseudosagittal triradiates. At present this genus is represented by two species only, the second of which being *A. ijimai*¹ from Japan and which was described by the present writer in 1916.

The present record reports the occurrence of *A. paulini* in Portugal for the second time.

Family Grantiidae DENDY

Genus GRANTIA FLEMING

11) *Grantia mexico*, n. sp.

(Pl. VII, Fig. 11; textfig. 8)

This new species is based upon a single specimen in the collection (Spec. No. 175₈).

The sponge is of a tubular form slightly curved, broadest at the attachment base and gradually narrowing towards the upper osculum which

¹*Amphiute ijimai*, HÔZAWA, 1916, pp. 33-38, pl. I, Fig. 9, Pl. II, Fig. 17; textfig. 7; 1929 p. 319.

is nearly naked. It is about 10 mm. long, and about 5 mm. broad at the base. The sponge wall is about 1 mm. thick. The osculum is about 1 mm. in diameter.

The dermal surface is more or less hispid due to the projecting oxea and the gastral surface is also echinated by the projecting apical rays of the gastral quadriradiates.

The colour in alcohol is greyish white and the texture is fairly firm.

Structure : - The canal system is of the syconoid type. The flagellated chambers are of elongated and 100-150 μ in diameter. Most of these chambers are simple not giving off any branches. The dermal skeleton is rather weakly developed, being made up of tangential triradiates in a few layers. The large oxea are placed mostly at right angles to the dermal surface, with their distal ends freely projecting on it, and with their proximal ends deeply intruding into the chamber layer. In addition to the large oxea above mentioned the hair-like spicules may be found in the dermal cortex. They are grouped in small tufts and are placed at right angles to the dermal surface.

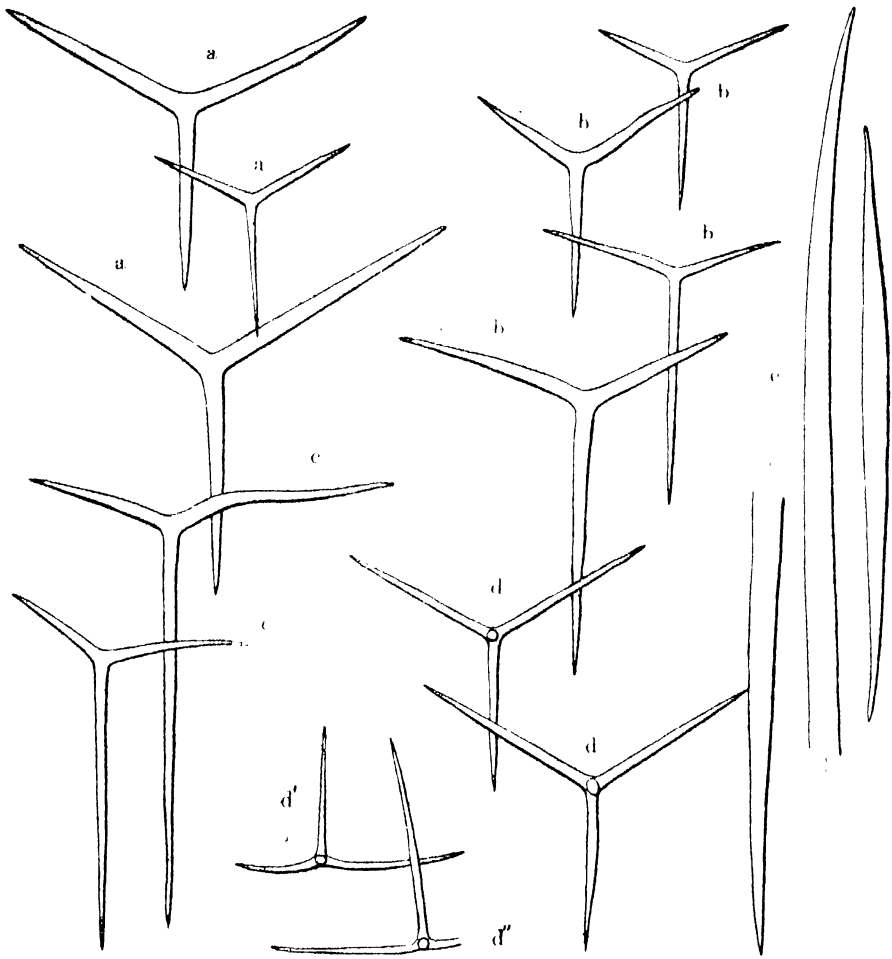
The tubar skeleton which is made up chiefly of triradiates is of the many-jointed articulate type. The inner most joint of the tubar skeleton is found by the basal rays of subgastral triradiates. The gastral skeleton consists of quadriradiates, of which the basal ray generally points toward the base of the sponge, and the apical ray projects into the gastral cavity and is curved towards the osculum. As regards the skeleton of the oscular margin, we have not noticed any peculiarities to be mentioned.

Spicules (textfig. 8). Dermal triradiates (a) slightly sagittal with paired rays slightly shorter and thicker than basal ray. All rays taper from base to sharp point. Basal ray straight, being 112-235 μ long by 6-14 μ thick. Paired rays slightly curved forwards, being 90-190 μ long by 8-16 μ thick.

Tubar triradiates (b) sagittal. Basal ray straight slightly longer than paired rays, not lying in the same plane with the latter, 112-225 μ long and 8-14 μ thick at the base. Paired rays curved around the flagellated chambers, 73-151 μ long and 8-14 μ thick at the base.

Subgastral triradiates (c) strongly sagittal. Rays almost lying in one plane. Basal rays, 240-300 μ long and 8-14 μ thick at the base. Paired rays widely diverging, distinctly curved at the point a little distant from the base, very sharply pointed, 84-201 μ long and 8-14 μ thick at the base.

Gastral quadriradiates (d) sagittal. Facial rays of subequal length and thickness lying in one plane. Basal ray straight, more or less shorter



Textfig. 8. *Grantia mexico*, n. sp. a, dermal triradiates; b, tubar triradiates; c, subgastral triradiates; d, gastral quadriradiates; d', gastral quadriradiate seen from above; d'', gastral quadriradiate seen from side; e, oxea (all $\times 150$).

than paired rays, $120\text{--}123\ \mu$ long and $8\text{--}10\ \mu$ thick at the base. Paired rays slightly curved backwards, $123\text{--}140\ \mu$ long and $8\text{--}10\ \mu$ thick at the base. Apical ray gently curved upwards, very sharply pointed, $110\text{--}173\ \mu$ long and $6\text{--}8\ \mu$ thick at the base.

Oxea projecting from the dermal surface (e) straight or slightly curved, sharply pointed at both ends. These spicules varies in length measuring from $526\ \mu$ up to $1110\ \mu$. In thickness they are $20\text{--}24\ \mu$.

Hair-like spicules projecting from the dermal surface very slender

and are easily broken into pieces. They are much shorter than the *oxea* above mentioned and are only 2μ thick.

Locality: — Mexico.

Remarks: — The above described species cannot be identified with any species previously described. *Grantia canadensis* LAMBE¹⁾ may be looked upon as its nearest ally though it shows some distinct differences in spiculation.

Genus *ANAMIXILLA* POLÉJAEFF

12) *Anamixilla irregularis* BURTON

(Pl. VII, Fig. 12)

Anamixilla irregularis, BURTON, 1930, p. 6, textfig. 5.

This species is represented in the Hamburg Museum collection by two specimens (Spec. Nos. 1110 a and b). They are in the form of several tubular individuals arising from a common basal mass. Some of them are provided with a naked osculum at each apex while the others are blind. In the larger specimen (Spec. No. 1110 a) the breadth of each individual varies 3-7 mm. and the thickness of the wall measures from 0.75 to 1 mm.

The writer has identified these two specimens with *Anamixilla irregularis* from Bay of Bima, Sumbawa, which was described by BURTON as they seem to agree very well in all essential characteristics with that species.

Localities: — Bay of Bima, Sumbawa (BURTON); Haiti.

Genus *LEUCANDRA* HAECKEL

13) *Leucandra pumila* BOWERBANK

(Pl. VII, Fig. 14, textfig. 9)

Leuconia pumila, BOWERBANK, 1866, p. 41; GRAY, 1867, p. 556.

Leucaltis pumila, HAECKEL, 1872, p. 148, Taf. 27, fig. 2 a-2 g.

Leucandra pumila, DENDY and ROW, 1913, p. 774.

This species is represented in the collection by a single specimen (Spec. No. 175₁) obtained from Mexico.

It is a solitary tubular individual more or less irregular in contour, being broadest near the attachment base and gradually narrowing towards the tip where an osculum opens. It measures about 14 mm. in total length and 9 mm. in the greatest breadth. The thickness of the wall is under

¹⁾*Grantia canadensis* LAMBE, 1896, pp. 206-207, pl. III, figs. 7 a, 7 b, 7 c.

1 mm. The osculum is about 0.5 mm. in diameter.

The colour is greyish white and the texture is fairly rigid.

Structure:— The canal system is of the leuconoid type. The flagellated chambers are rather thinly distributed in the chamber layer and are irregularly scattered. They are generally of an oval shape, measuring up to 120μ in the longer diameter.

The dermal skeleton is composed chiefly of rather thin triradiates which are tangentially but confusedly arranged in a few layers and here may be added a number of large triradiates which are placed again tangentially. The tubar skeleton is made up chiefly of large triradiates in an irregular arrangement. The quadriradiates which occur along the larger exhalant canals may be added to the skeleton. Their apical rays project into the lumen of the canal.

In the subgastral position there occur a small number of triradiates and quadriradiates with their basal rays intruding into the chamber layer.

The gastral skeleton is made up of quadriradiates in a few layers with the basal rays pointing downwards in most cases and with apical rays projecting into the gastral cavity. Some of these quadriradiates become triradiates being deprived of their apical ray.

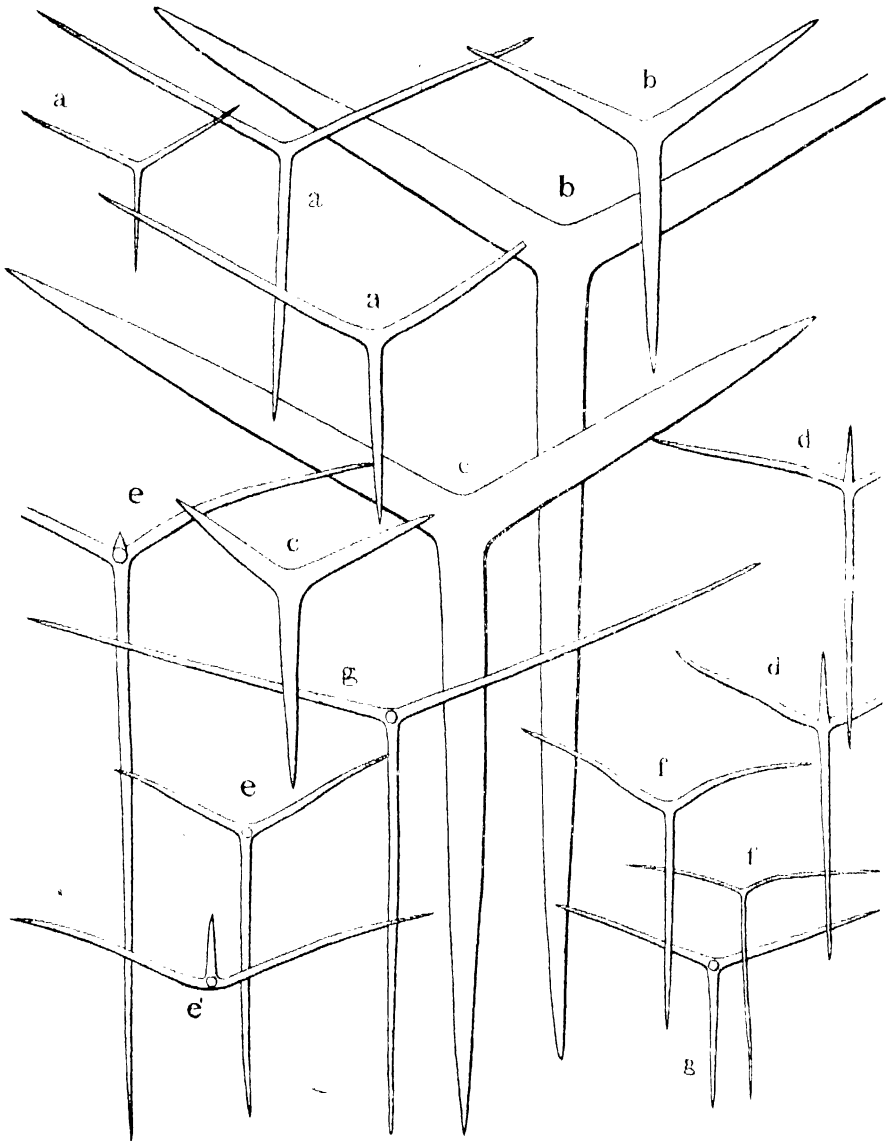
As regards the skeleton of the oscular margin we have not noticed any peculiarities to be mentioned.

Spicules (textfig. 9):— Small dermal triradiates (a) slightly sagittal with paired rays slightly longer than basal ray. All rays are of equal thickness and taper from base to sharp point. Basal ray straight, being $130\text{--}325\mu$ long by $8\text{--}16\mu$ thick. Paired rays slightly curved forwards, being $160\text{--}390\mu$ long by $8\text{--}16\mu$ thick.

Larger dermal triradiates (b) equiradiate and equiangular, or slightly sagittal. All rays straight, gradually and sharply pointed, being $250\text{--}1,000\mu$ long and $30\text{--}60\mu$ thick at the base.

Tubar triradiates regular or subregular (c), very large but variable in size. They are exactly similar to the larger triradiates found in the dermal cortex.

Quadriradiates of the larger exhalant canals (d) sagittal. Basal ray straight not strongly differentiated in length from paired rays, being $170\text{--}490\mu$ long and $12\text{--}14\mu$ thick at the base. Paired rays usually equal, sometimes unequal, either curved simply forwards or showing double curvature. In the latter case they curve distinctly forward in the proximal parts and slightly backwards in the distal parts, $200\text{--}480\mu$ long and $12\text{--}14\mu$ thick at the base. Apical ray much shorter and slightly thinner than



Textfig. 9. *Leucandra pumila* BOWERBANK. a, small triradiates; b, larger dermal triradiates; c, tubar triradiates; d, quadriradiates of larger exhalant canal; e, subgastral quadriradiates; e', the same seen from above; f, subgastral triradiates; g, gastral quadriradiates (all $\times 100$).

facial rays, slightly curved and finely pointed, $110\text{--}140\ \mu$ long and about $12\text{--}14\ \mu$ thick at the base.

Subgastral quadriradiates (e) strongly sagittal. Basal ray much longer than paired rays, being about $340\text{--}780\ \mu$ and $14\text{--}18\ \mu$ thick at the base. Paired rays widely diverging and slightly curved, about $190\text{--}340\ \mu$ long and $12\text{--}16\ \mu$ thick at the base. Apical ray short and nearly straight, broad at the base, and very finely pointed at the end, being about $50\text{--}90\ \mu$ long and $12\text{--}16\ \mu$ thick at the base.

Subgastral triradiates (f) strongly sagittal. They are nearly similar to subgastral quadriradiates except in the absence of an apical ray.

Gastral quadriradiates (g) sagittal. Basal ray not strongly differentiated in length from paired rays, quite straight, being $170\text{--}490\ \mu$ long and $12\text{--}14\ \mu$ thick at the base. Paired rays nearly straight or very slightly curved, rather widely diverging, sharply pointed, being about $200\text{--}408\ \mu$ long and $12\text{--}14\ \mu$ thick at the base. Apical ray fairly well-developed, shorter than facial rays, slightly curved upwards and sharply pointed, measuring $110\text{--}140\ \mu$ in length and $12\text{--}14\ \mu$ broad at the base.

Previously known Distribution: --- Guernsey (BOWERBANK); Mogador, Coast of Morocco; Cape of Good Hope; Bass Strait, Indian Ocean (HAECKEL).

Locality: Mexico.

Remarks: -- This species was first described by BOWERBANK in 1886, who obtained the specimen from Guernsey in the English Channel. Afterwards it was more fully reported by HAECKEL in 1872 in his Monograph of Calcareous, the specimens being secured from the Coast of Morocco, Cape of Good Hope, and from the Bass Strait in the Indian Ocean. This time the specimen was obtained from Mexico.

As the present specimen from Mexico coincides well in external form, as well as in skeletal features with those mentioned by BOWERBANK and HAECKEL in the case of *Leucandra pumila*, the writer has identified it with the present species.

In the present report the writer has mentioned the existence of subgastral triradiates and of subgastral quadriradiates in this species.

14) *Leucandra seychellensis*, n. sp.

(Pl. VII, Fig. 13; textfig. 10)

This species is represented by a single specimen in the collection.

The sponge is a solitary individual of an irregular oval shape. It measures about 30 mm. in height and about 22 mm. in the greatest breadth, the wall being about 5 mm. in thickness. The osculum at the upper end is rather irregular in outline and is surrounded by a thin margin. It

measures about 6 mm. across. The dermal surface is nearly smooth without any projecting spicules, but is more or less uneven as there exist a number of protuberances and depressions distributed at intervals. The gastral surface is also smooth without any strongly projecting spicules and is perforated by irregularly distributed circular or oval exhalant apertures measuring up to 3 mm. in diameter.

The colour in alcohol is greyish white and the texture is moderately firm and elastic.

Structure :— The canal system is of the leuconoid type. The dermal cortex is very thin and underneath which there exist subdermal cavities. The water comes into these cavities from outside through the inhalant pores of dermal cortex and then is lead into the chamber layer. The chamber layer is strongly lacunar owing to the wide inhalant and exhalant canals. Between the inhalant and exhalant canal systems the flagellated chambers are fairly thickly distributed. They are ovoid or spherical in form with a diameter of about 100μ .

The dermal skeleton is very thin, composed of a few layers of small triradiates which are arranged without any definite orientation, but to form small reticular meshes.

In addition to these spicules there occur a number of colossal oxea. The arrangement of these spicules, except for the fact that they all lie tangentially, is quite irregular.

The skeleton of the chamber layer is rather confused, being composed chiefly of triradiates with an admixture of a few number of quadriradiates. The quadriradiates occur along the larger exhalant canal.

The gastral skeleton is made up of a thin layer of triradiates which are closely set without any definite orientation.

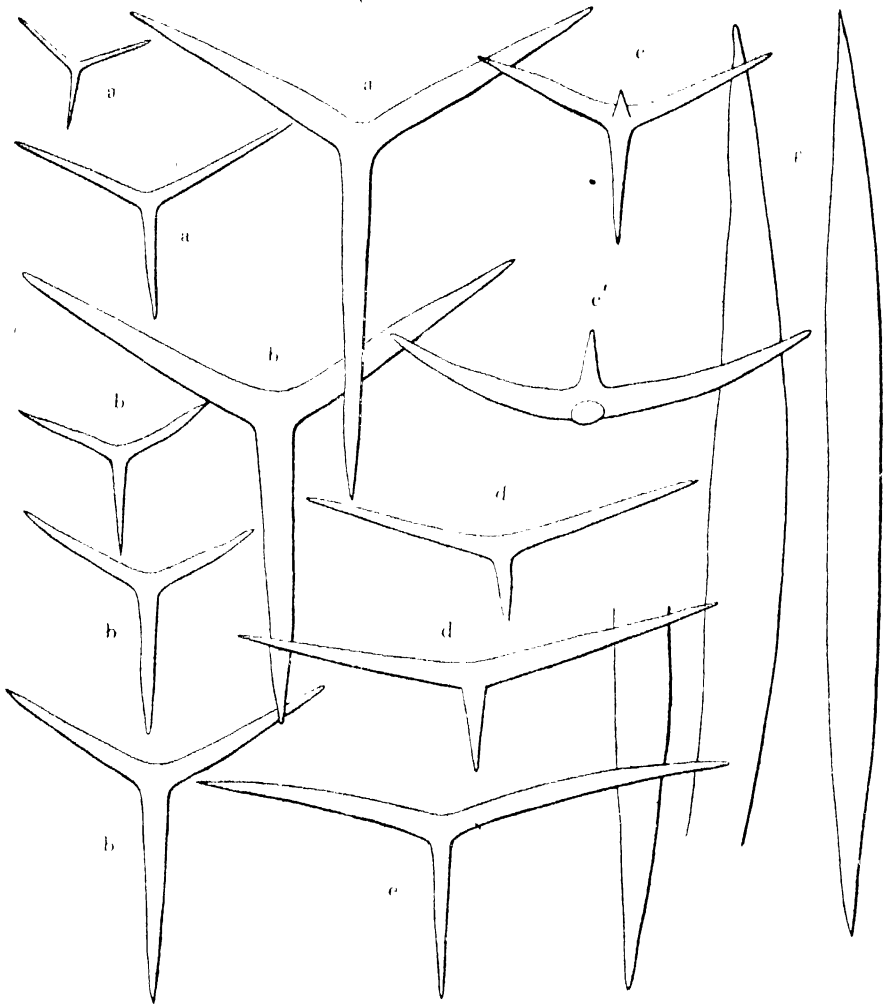
The skeleton of the oscular margin is composed of triradiates and colossal oxea. Of the triradiates, those found on the dermal surface are placed rather irregularly, while those on the gastral surface have their basal rays directed regularly downwards and running parallel with one another. The colossal oxea are disposed longitudinally and run parallel with one another.

Spicules (textfig. 10) :— Dermal triradiates (a) subregular or slightly sagittal with rays sharply pointed and varying fairly greatly in size. In the smaller spicules the rays measure about 70μ by 6μ while in the larger ones they are about 260μ by 20μ at the base.

Triradiates of the chamber layer (b) are nearly similar to the larger triradiates of the dermal cortex. They are slightly sagittal, the oral angle

being a little wider than the paired angles. All rays are nearly equally long and thick, and are sharply pointed at end. Basal ray straight, 130–230 μ long and 20–30 μ thick at the base. Paired rays slightly curved forwards, 140–240 μ long and 20–30 μ thick at the base.

Quadriradiates of the larger exhalant canal (c) like the triradiates of the chamber layer except for the presence of an apical ray. The apical



Textfig. 10. *Leucandra seychellensis*, n. sp. a, dermal triradiates; b, triradiates of chamber layer; c, quadriradiate of the layer exhalant canal; c', the same seen from above; d, gastral triradiates; e, triradiates of the oscular margin; f, dermal oxea (all $\times 150$).

ray is much shorter and thinner than facial rays and is very slightly curved ending in a sharp point, about 50μ long and about 14μ thick at the base.

Gastral triradiates (d) strongly sagittal having a very wide oral angle. Basal ray straight and sharply pointed, much shorter and slightly thinner than paired rays, $50-90\mu$ long and $6-8\mu$ thick at the base. Paired rays nearly straight and sharply pointed at the end, $130-180\mu$ long by $8-10\mu$ thick at the base.

Triradiates of the oscular margin (e) strongly sagittal. Basal ray distinctly shorter and thinner than paired rays. It is straight and sharply pointed and measures about 140μ long by $12-18\mu$ thick at the base. Paired rays very widely divergent and curve gently backwards, about 230μ long and $18-20\mu$ thick at the base.

Dermal oxea (f) very large, more or less curved, spindle shaped, the broadest portion lying nearer one end than the other, sharply pointed at both ends, $800-1050\mu$ long and $45-65\mu$ thick at the thickest portion.

Locality: Seychelle Islands, Indian Ocean.

Remarks: The most characteristic feature of this new species is the presence of colossal oxea in the dermal cortex. They are not arranged longitudinally, but lie scattered quite irregularly over the sponge surface and never project from the latter. The same feature is also seen in the cases of *Leucandra crambessa*¹ and its varieties which on the other hand differ from the species now described in the external form, in the features of the gastral skeleton, as well as in other respects. The gastral skeleton of *Leucandra crambessa* and its varieties is composed chiefly of quadri-radiates, while in the species now described it is made up almost entirely of triradiates.

LIST OF REFERENCES

- BOWERBANK, J. S. (1866). A Monograph of the British Spongiadae. Vol. II. Ray. Soc. London.
- BREITFUSS, L. (1896). Kalkschwämme der Bremer-Expedition nach Ost-Spitzbergen im Jahre 1889. Zool. Anzeiger, Bd. XIX, No. 514, s. 426-432.
- (1898). Die Kalkschwammfauna von Spitzbergen. Nach den Sammlungen der Bremer-Expedition nach Ost-Spitzbergen im Jahre 1889 (Prof. W. KÜENTHAL und Dr. A. WALTER). Zoolog. Jahrbuch, Syst. Abth., Bd. XI, s. 103-120, Taf. 12, 13.
- (1935). Le Spugne calcaree dell' Adriatico con riflesso a tutto il Mediterraneo. Consiglio Nazionale delle Ricerche R. Comitato Talassografico Italiano, Memoria CCXXIII.

¹*Leucandra crambessa*, HAECKEL, (1872), p. 182, Taf. 32, fig. 3 a-g; Taf. 37, fig. 7A, B, 8.

- BURTON, M. (1930). The Porifera of the Siboga Expedition III. Calcareae.
- CARTER, H. J. (1886). Descriptions of Sponges from the Neighbourhood of Port Phillip Heads, South Australia. *Ann. Mag. Nat. Hist.*, (Ser. 5), Vol. XVII, pp. 502-516.
- DENDY, A. (1892). Synopsis of the Australian Calcareae Heterocoela, with a Proposed Classification of the Group, and Descriptions of Some New Genera and Species. *Proc. Roy. Soc. Victoria* (n. s.), Vol. V, pp. 69-116.
- (1893). Studies on the Comparative Anatomy of Sponges. V. Observations on the Structure and Classification of the Calcareae Heterocoela. *Quart. Journ. Microsc. Sci.*, (n. s.), Vol. XXXV, pp. 159-257.
- (1905). Report on the Sponges collected by Professor HERDMAN at Ceylon in 1902. *Reports on the Pearl Oyster Fisheries of the Gulf of Manaar*, Vol. III, pp. 59-246.
- (1913). Report on the Calcareous Sponges collected by the Sealark Expedition in the Indian Ocean. *Trans. Linnean Soc. London, Zool.* Vol. XVI, pp. 1-29.
- DENDY, A. and ROW, R. W. H. (1913). The Classification and Phylogeny of the Calcareous Sponges, with a Reference List of all the Described Species, Systematically Arranged. *Proc. Zool. Soc. London*, 1913, pp. 704-813.
- ELLIS, J. and SOLANDER, D. (1786). Natural History of Many Curious and Uncommon Zoophytes collected from Various Parts of the Globe. London, 1786.
- GRAY, J. E. (1867). Notes on the Arrangement of Sponges, with the Description of Some New Genera. *Proc. Zool. Soc., London*, 1867, pp. 492-558, pls. XXVII, XXVIII.
- HAECKEL, E. (1870). *Prodromus eines Systems der Kalkschwämme*. *Jenaische Zeitschr.*, Bd. V, s. 236-254.
- (1872). *Die Kalkschwämme, eine Monographie*. Berlin.
- HANITSCH, R. (1895). Notes on a Collection of Sponges from the West Coast of Portugal. *Trans. Liverpool Biol. Soc.*, Vol. IX, pp. 205-219.
- HÔZAWA, S. (1916). On Some Japanese Calcareous Sponges belonging to the Family Heteropiidae. *Journ. Coll. Sci. Imp. Univ. Tôkyo*, Vol. XXXVIII, Art. 5, pls. I, II.
- (1918). Report on the Calcareous Sponges collected by the U. S. Fisheries Steamer "Albertross" in the Northwestern Pacific during 1906. *Proc. U. S. Nat. Mus.*, Vol. 54, pp. 525-556, pl. 84, 85, 1918.
- (1929). Studies on the Calcareous Sponges of Japan. *Journ. Fac. Sci. Imp. Univ. Tôkyo*, Sec. IV. Zoology, Vol. 1, Part 5, pp. 277-389, pl. I-XII.
- (1933). Report on the Calcareous Sponges obtained by the Survey of the Continental Shelf bordering on Japan. *Science Reports Tôhoku Imp. Univ.*, (Ser. IV) Biology, Vol. VIII, No. 1, 1933, pp. 1-20, pl. I.
- JENKIN, C. F. (1908A). The Calcareous Sponges (in) *The Marine Fauna of Zanzibar and British East Africa*, from Collections made by Cyrill Crossland, MA. in the Years 1901 and 1902. *Proc. Zool. Soc. London*, 1908, pp. 434-456.
- (1908B). The Calcareae of the National Antarctic Expedition. *National History Reports*, Vol. IV.
- LACKSCHWITSCH, P. (1886). Über die Kalkschwämme Menorcas. *Zool. Jahrbuch*, Bd. I, S. 291-310.
- LAMBE, L. M. (1896). Sponges from the Atlantic Coast of Canada. *Trans. Roy. Soc. Canada* (Ser. 2), Vol. II, pp. 181-211.
- LENDENFELD, R. VON. (1885). A Monograph of the Australian Sponges. Part III. The Calcispongiae. *Proc. Linn. Soc. New South Wales*, Vol. IX, pp. 1083-1150.

- LIEBERKÜHN, N. (1859). Neue Beiträge zur Anatomie der Spongien. MÜLLER's Archiv, 1859, S. 353-382, 515-530.
- MICHLUCHO-MACLAY, N. DE. (1868). Beiträge zur Kenntniss der Spongien. Jenaische Zeitschr., Bd. IV, S. 221-240.
- MINCHIN, E. A. (1905). The Characters and Synonymy of the British Species of Sponges of the Genus *Leucosolenia*. Proc. Zool. Soc., London, 1904, Vol. II, pp. 349-396.
- MONTAGU, G. (1812). An Essay on Sponges, with Descriptions of all the Species that have been discovered on the Coast of Great Britain. Memoirs of the Wernerian Soc. Edinburg, Vol. II, pp. 67-122.
- POLÉJAEFF, N. (1883). The Calcarea. Report on the Scientific Results of H. M. S. 'Challenger', Zoology, Vol. VIII.
- RIDLEY, S. O. (1884). Spongida. Reports on the Zoological Collections made in the Indo-Pacific Ocean during the Voyage of H. M. S. 'Alert', 1881-1882, pp. 366-482, 582-630, London, 1884.
- SCHMIDT, O. (1862). Die Spongien des Adriatischen Meeres. Leipzig, 1862.
- SCHUFFNER, O. (1877). Beschreibung einiger neuer Kalkschwämme. Jenaische Zeitschr. Bd. XI, S. 403-433, Taf. XXIX-XXVI.
- THACKER, A. G. (1908). On Collections of the Cape Verde Islands Fauna made by Cyril Crossland, 1904, the Calcareous Sponges. Proc. Zool. Soc. London, 1908, pp. 757-782.

EXPLANATION OF THE PLATES

PLATE VI.

- Fig. 1. *Leucosolenia complicata* (MONTAGU). ×2.
- Fig. 2. *Leucosolenia canariensis* MICHLUCHO-MACLAY. ×2.
- Fig. 3. *Leucaltis clathria* HAECKEL. ×2.
- Fig. 4. *Sycon mexicanum*, n. sp. ×2.
- Fig. 5. *Sycon coronatum* ELLIS et SOLANDER. ×2.
- Fig. 6. *Vosmaeropsis japonica* HÔZAWA. ×2.
- Fig. 7. *Vosmaeropsis simplex*, n. sp. ×1.

PLATE VII.

- Fig. 8. *Vosmaeropsis levis*, n. sp. ×2.
- Fig. 9. *Vosmaeropsis triradiata*, n. sp. ×2.
- Fig. 10. *Amphiute paulini* HANITSCH. ×2.
- Fig. 11. *Grantia mexicana*, n. sp. ×2.
- Fig. 12. *Anamixilla irregularis*, n. sp. ×2.
- Fig. 13. *Leucandra seychellensis*, n. sp. ×2.
- Fig. 14. *Leucandra pumila* BOWERBANK. ×2.



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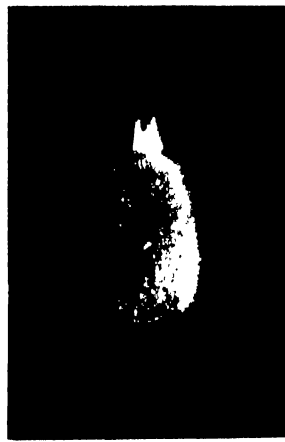
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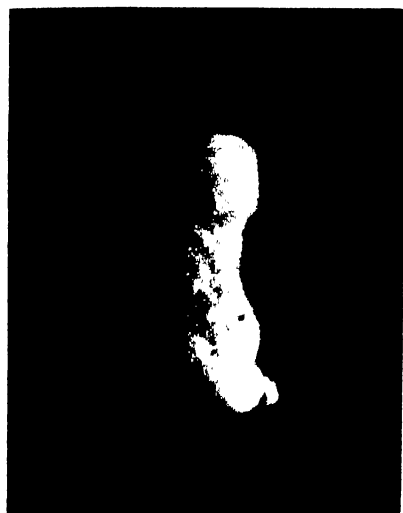
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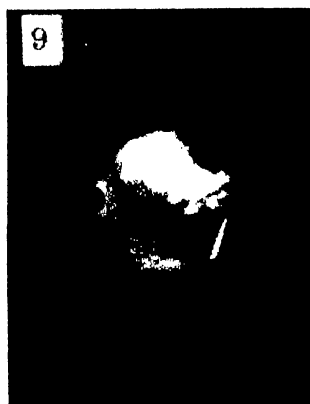
S. HOZAWA: Calcareia obtained by the Hamburg Museum.



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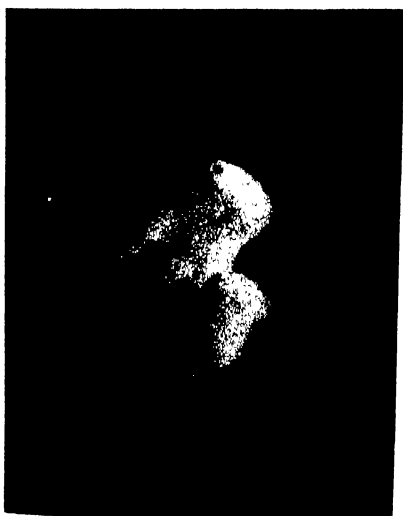
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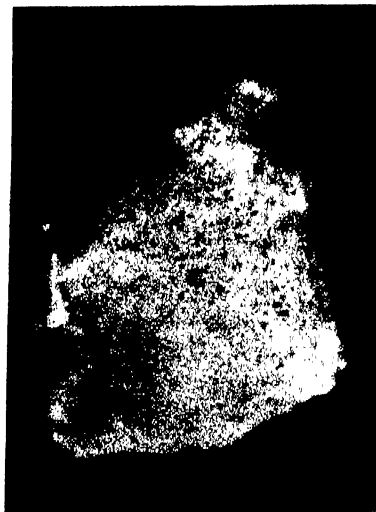
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S. HOZAWA: Calcarea obtained by the Hamburg Museum.

CALCAREOUS SPONGES OF MATSUSHIMA BAY

By

SENJI TANITA

Biological Institute, Tôhoku Imperial University, Sendai, Japan

(With Plate VIII and 4 text-figures)

(Received February 1, 1940)

The calcareous sponges reported in the present paper are those collected from Matsushima Bay by Professor HOZAWA, by Dr. URAGAMI and by the writer during the years extending from 1935 to 1939. The collection was found to comprise six species listed below, of which four seem to be new to science.

Before going further, the writer wishes to acknowledge his indebtedness to Professor HOZAWA for the valuable advice and kind help rendered him during the work. The writer also wishes to thank Dr. URAGAMI for his collection of material offered for use.

The following is the list of the species.

Family Homocoelidae

- 1) *Leucosolenia mutsu* HOZAWA
- 2) *Leucosolenia tenera*, n. sp.

Family Sycettidae

- 3) *Sycon okadai* HOZAWA
- 4) *Sycon matsushimense*, n. sp.
- 5) *Sycon uragamii*, n. sp.

Family Grantiidae

- 6) *Leucandra tomentosa*, n. sp.

DESCRIPTION

1. *Leucosolenia mutsu* HOZAWA

(Pl. VIII, fig. 1)

Leucosolenia mutsu HOZAWA, 1928, pp. 219-220, Pl. I. figs. 1-3; 1940, p. 35.

This species is represented in the collection by two specimens obtained from the littoral zone of Mahanashi-jima in 1939.

The sponges form irregular spreading masses, each consisting of a loose network of Ascon-tubes. One of them attains the length of 12 mm.

and the other of 4 mm. only.

With respect to the inner structure, these specimens are identical with the type.

Localities: --- Mutsu Bay; Ohshima in Kesennuma Bay (HOZAWA); Mahanashi-jima in Matsushima Bay.

2. *Leucosolenia tenera*, n. sp.

(Pl. VIII, fig. 2; Textfig. 1)

Many specimens of this new species exist in the collection. Some of them were obtained by Professor HOZAWA in 1938 on the shore of Mahanashi-jima, and some others were collected by the writer on the same island in 1939.

The sponge forms a loose, branching mass of Ascon-tubes of variable sizes and shapes, attached to some foreign object. The sponge seems to increase its size by budding. The bud which first appears on the parent tube as a blind outgrowth, comes subsequently to bear an osculum at its free end. The ascon-pores are small, in the form of thin-walled tubes, each when fully grown bearing a circular osculum of about 1.2 mm. diameter at its extremity. The length and the diameter of the tubes are variable corresponding to the position taken in the colonies, measuring about 3–12 mm. in length and 0.5–3 mm. in diameter.

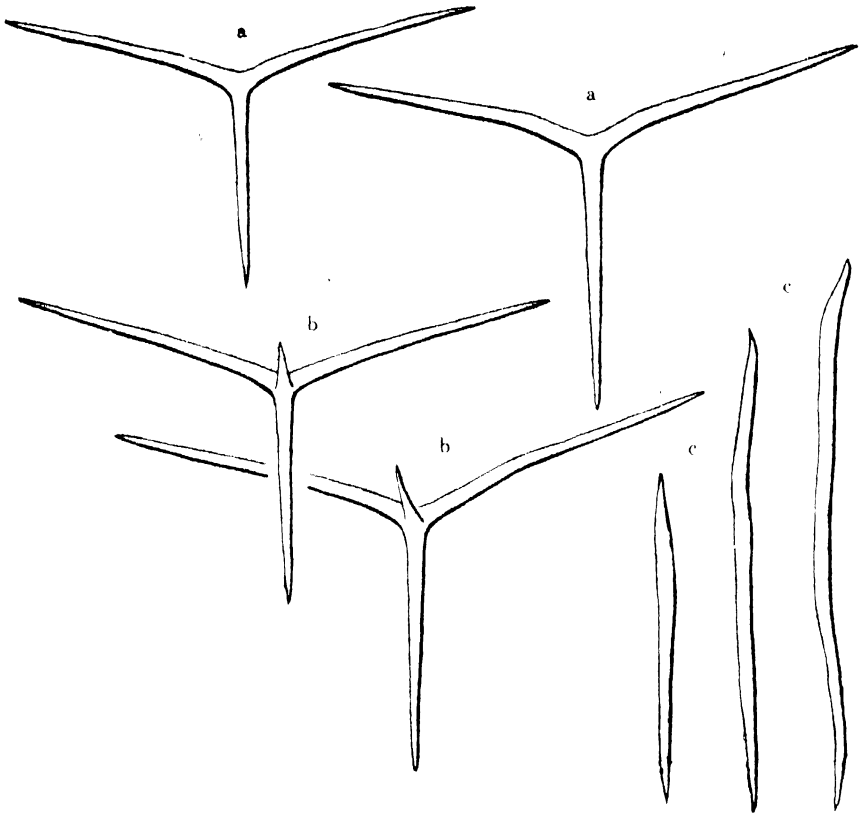
The outer surface of the tubes is very minutely hispid. The colour in alcohol is nearly white and the texture is soft and bristle.

Structure: --- The canal system is of DENDY's type *simplicia*¹. The skeleton consists of triradiates, quadriradiates and oxea. The triradiates and quadriradiates are arranged in a thin layer, the apical rays of the latter projecting into the gastral cavity. The inner end of the oxea is embedded in the mesoderm while the outer projects outwards giving to the outer surfaces of the tubes the hispid appearance.

Spicules (Textfig. 1): --- Triradiates (a) sagittal. All rays are nearly equal in thickness being 7–10 μ . The basal ray straight, gradually tapering to a sharp point, always shorter than paired rays being 80–180 μ long. Paired rays nearly equal, widely divergent, straight or slightly curved backwards and are 90–210 μ long.

Quadriradiates (b) are of about the same size and shape as the triradiates with addition of an apical ray. The apical ray slightly curved upwards, sharply pointed, shorter and thinner than the facial rays, 30–

¹DENDY, A. Trans. Roy. Soc. Victoria, Vol. III, 1891, p. 24.



Text-fig. 1. *Leucosolenia tenera*, n. sp. a, triradiates; b, quadriradiates; c, oxea. (all $\times 200$)

$70\ \mu$ long and $6-8\ \mu$ thick at base.

Oxea (c) more or less irregularly curved, sharply pointed at both ends, but the distal end is usually slender and is more finely pointed than the proximal. They are not even in outline, $200-530\ \mu$ long and $8-12\ \mu$ thick at the broadest portion.

Remarks:—In the external features and canal system, this species closely resembles *Leucosolenia lucasi* DENDY¹ and *L. echinata* KIRK², but it may be easily distinguished from them by its spiculation. In the present species the radiates are more widely divergent and the basal rays are always shorter than paired rays. But in *L. lucasi*, the three angles of the triradiates are equal, and in *L. echinata* the triradiates are regular

¹ *Leucosolenia lucasi* DENDY, (1891), pp. 45-46, Pl. I, fig. 1, Pl. IV, fig. 1, Pl. IX, fig. 1.

² *Leucosolenia echinata* KIRK, (1893), pp. 177-178, Pl. XXII, fig. 1.

or sagittal and the quadriradiates are larger than the triradiates. Moreover the present species may be distinguished from them by the difference in the form of the oxeote spicules.

Locality: — Mahanashi-jima in Matsushima Bay.

3. *Sycon okadai* HOZAWA

(Pl. VIII, fig. 3)

Sycon okadai HOZAWA, 1929, pp. 302-304, Pl. III, figs. 18, 19, textfig. 10.

This species is represented by numerous specimens in the collection. One of them (Pl. VIII, fig. 3) was obtained by Professor HOZAWA from the shore of Mahanashi-jima, while the others were collected by Dr. URAGAMI from the oyster beds in Matsushima Bay. All are typical, forming cylindrical solitary persons. The largest specimen measures about 27 mm. in total length and about 7 mm. in the greatest breadth, the wall reaching about 1.2 mm. in thickness. The osculum at the upper end is oval in shape and is surrounded by a feebly developed collar. The remaining specimens are much smaller than the specimen above alluded to.

In the anatomical structures, present specimens represent the same features with the type which was first described by HOZAWA, using specimens from Misaki.

Localities: — Misaki (HOZAWA); Matsushima Bay.

4. *Sycon matsushimense*, n. sp.

(Pl. VIII, fig. 4, Textfig. 2)

This new species is based upon five specimens found in the collection. They were collected by the writer at the littoral zone of Mahanashi-jima in Matsushima Bay in May, 1939. The sponge has the form of an elongate sac with a circular osculum surrounded by a well-developed collar at the upper end and is attached directly with its base to other subjects. The largest specimen which I have selected as the type is about 18 mm. in total length and 4.5 mm. in breadth. The osculum measures about 2.5 mm. in diameter and the height of the collar is 5 mm. The body wall of the sponge reaches 1.5 mm. in thickness in the thickest parts.

The dermal surface is fairly hispid owing to the oxea projecting from it. The gastral surface appears slightly rough under the microscope, due to the projecting apical rays of the gastral quadriradiates.

The colour in alcohol is greyish white and the texture is soft but

slightly elastic.

Structure: - The canal system is syconoid. Flagellate chambers are cylindrical in form, straight, unbranched, and are rounded at the slightly projecting ends. They are $160-200\mu$ in diameter while the length are variable corresponding to the thickness of sponge wall.

The tubar skeleton consists of triradiates arranged in several layers with outwardly directed basal rays and of basal rays of subgastral tri- and quadriradiates. The distal ends of flagellate chambers are provided each with a tuft of oxea of three kinds, viz. ordinary oxea, linear spicules and hair-like spicules. The ordinary oxea are several in number while the linear spicules are only 2 or 3.

The gastral skeleton is made up of paired rays of subgastral tri- and quadriradiates and of gastral quadriradiates with their basal rays mostly pointed towards the sponge base. The subgastral quadriradiates are very few in number in comparison with the subgastral triradiates.

The oscular collar consists of linear spicules and of quadriradiates. The former occur in longitudinal disposition at the distal end of collar forming a well-developed fringe. The latter spicules have their basal rays running parallel with the former kind of spicules and their paired rays widely divergent.

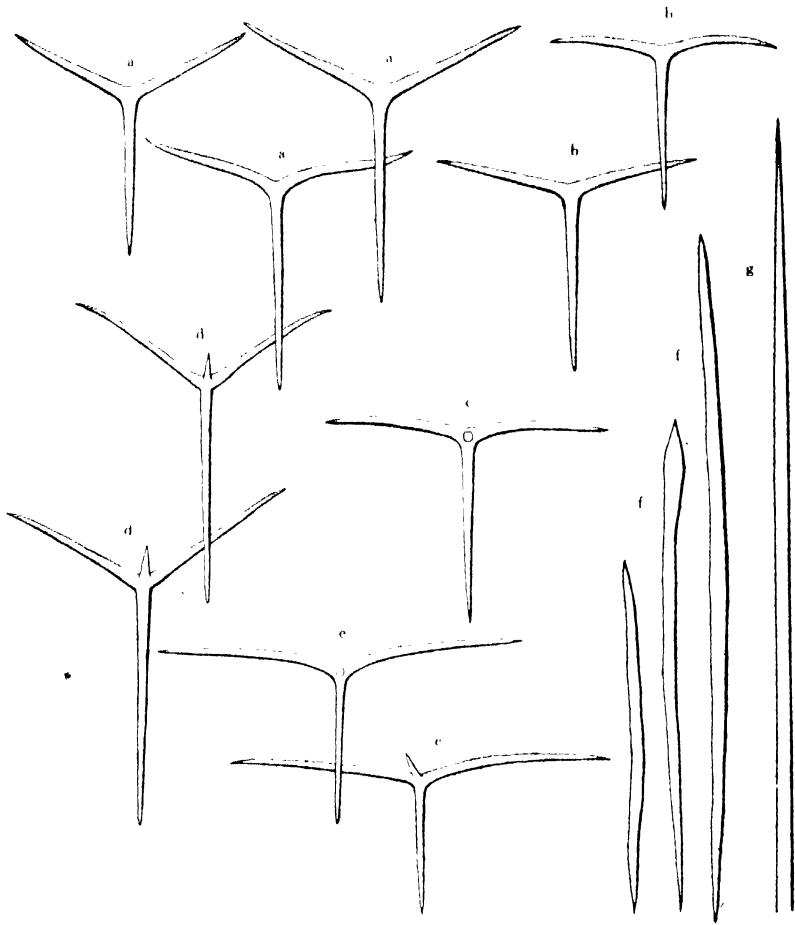
Spicules (Textfig. 2): Tubar triradiates (a) sagittal. All rays are nearly equal in thickness. Basal ray straight, sharply pointed, slightly longer than paired rays, $180-270\mu$ long and $8-10\mu$ thick at base. Paired rays equal, either straight or slightly curved forwards, $120-190\mu$ long and $8-10\mu$ thick at base.

Subgastral triradiates (b) sagittal. Basal ray straight, longer than paired rays, $190-220\mu$ long and $8-10\mu$ thick at base. Paired rays equal, widely divergent, $150-200\mu$ long and $8-10\mu$ thick at base.

Subgastral quadriradiates (c) similar to the subgastral triradiates, except the presence of apical rays. Apical ray slightly curved, sharply pointed, shorter and slightly thinner than facial rays, $60-90\mu$ long and about 7μ thick at base.

Gastral quadriradiates (d) strongly sagittal. Basal ray straight, distinctly longer than paired rays, $220-260\mu$ long and $8-10\mu$ thick at base. Paired rays nearly equal, slightly curved backwards, $140-190\mu$ long and $8-10\mu$ thick at base. Apical ray curved upwards, shorter and thinner than facial rays, $65-100\mu$ long and $6-8\mu$ thick at base.

Quadriradiates of the oscular margin (e) strongly sagittal. Basal ray straight, sharply pointed, shorter and slightly thinner than paired rays,



Textfig. 2. *Sycon matsushimense*, n. sp. a, tubar triradiates; b, subgastral triradiates; c, subgastral quadriradiate; d, gastral quadriradiates; e, quadriradiates of oscular margin; f, oxea at the distal end of flagellate chamber, g, linear spicule (all $\times 120$)

160–220 μ long and about 8 μ thick at base. Paired rays equal, very widely divergent, slightly curved backwards, 230–320 μ long and 8–11 μ thick at base. Apical ray short, curved upwards, about 70 μ long and 8 μ thick at base.

Oxea at the distal end of flagellate chambers (f) are more or less curved, tapering to both ends, not even in outline, 420–850 μ long and 14–18 μ thick in the middle. Some oxea are provided with an indistinct lance-head at distal end, while the others are equally sharply pointed at

both ends.

Linear spicules of the distal end of flagellate chambers (g) very slender, straight, uniformly thick throughout the entire length except for both sharply pointed ends, about 3.6 mm. long and $10\ \mu$ thick.

Hair-like oxea very fine, more or less curved, reaching a length of 2 mm. by $2\ \mu$ thick.

Linear spicules of oscular collar similar to those found at the distal end of flagellate chambers. They are straight, slender, variable in thickness, about 5.5 mm. long and $5\text{--}10\ \mu$ thick.

Locality: --- Mahanashi-jima in Matsushima Bay.

5. *Sycon uragami*, n. sp.

(Pl. VIII, fig. 5; Textfig. 3)

This species is based upon six specimens which were obtained by Dr. URAGAMI from the oyster beds in Matsushima Bay in October, 1935. They are all of a closely similar appearance. Each of them represents a solitary person of a tubular form, attached with a hollow peduncle to some foreign object and showing at the upper end an osculum surrounded by a feebly developed collar.

The largest specimen (Pl. VIII, fig. 5) which is herewith taken as the type, measures 9 mm. in total length and 2 mm. in the greatest breadth. The peduncle of the sponge is about 3 mm. long and the osculum is circular with a diameter of 1 mm. The sponge wall is about 0.5 mm. thick in the middle parts of the body.

The dermal surface of the sponge is hispid owing to the projecting oxea. The gastral surface appears nearly smooth to the naked eye. The gastral cavity extends the entire length of the sponge body. The peduncle is also tubular, its cavity communicating with the gastral cavity.

The colour in alcohol is greyish white and the texture is soft.

Structure: — Canal system typical. The flagellate chambers straight, unbranched, nearly equally thick throughout their entire length terminating in low rounded distal cones.

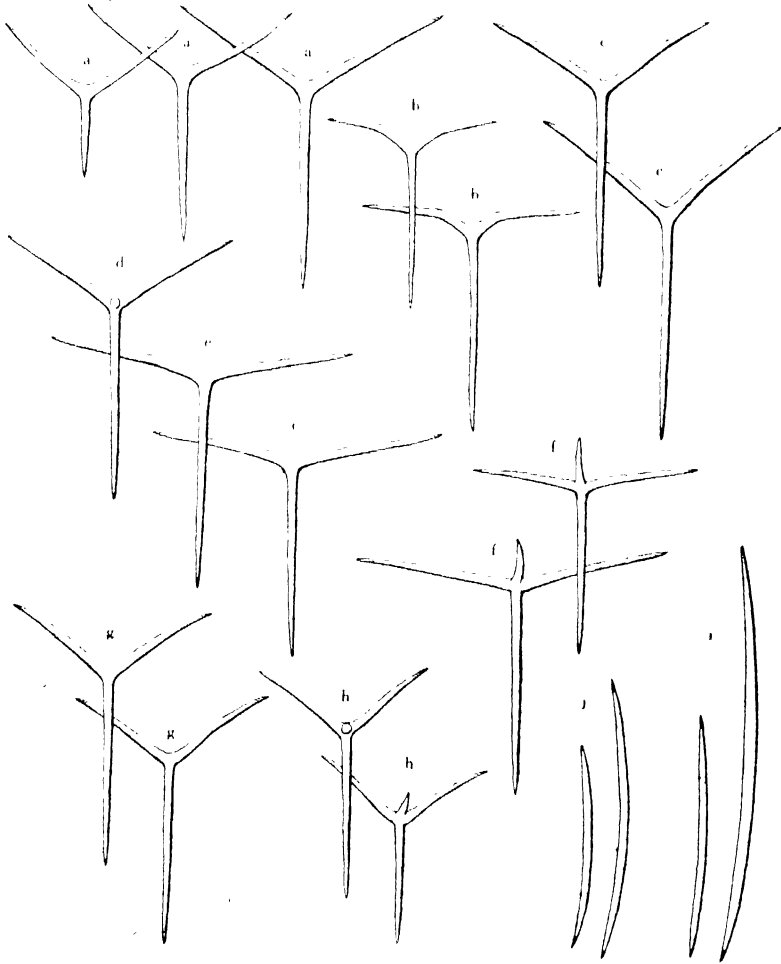
The tubar skeleton is composed of basal rays of subgastral triradiates and of articulate tubar triradiates. At the distal end of flagellate chamber occur the tufts of oxea.

The gastral skeleton is made up of paired rays of subgastral triradiates, gastral triradiates, and of gastral quadriradiates which have their apical rays projecting into the gastral cavity. The latter two kinds of spicules

are placed tangentially and their basal rays are pointed towards the sponge base.

The oscular margin is composed of linear spicules and of closely set triradiates and quadriradiates. The skeleton of the peduncle is consisted of tangentially placed triradiates and quadriradiates with their apical rays projecting into the cavity, and of radially arranged oxea.

Spicules (Textfig. 3): - Tubar triradiates (a) sagittal. Basal ray



Textfig. 3. *Sycon uragamii*, n. sp. a, tubar triradiates; b, subgastral triradiates; c, gastral triradiates; d, gastral quadriradiate; e, triradiates of oscular margin; f, quadriradiates of oscular margin; g, triradiates of peduncle; h, quadriradiates of peduncle; i, oxea of flagellate chamber; j, oxea of peduncle. (all $\times 120$)

straight, tapering to sharp end, longer than paired rays, 90–210 μ long and 6–9 μ thick at base. Paired rays equal, either straight or slightly curved forwards, 80–160 μ long and 6–9 μ thick at base.

Subgastral triradiates (b) strongly sagittal. All rays are nearly equally thick being 6–8 μ . Basal ray straight, longer than paired rays being 180–200 μ long. Paired rays equal, curved distinctly about in the middle and are 105–200 μ long.

Gastral triradiates (c) sagittal. Basal ray straight, sharply pointed, 170–250 μ long and 7–8 μ thick at base. Paired rays equal, nearly straight, 150–170 μ long and 7–8 μ thick at base.

Gastral quadriradiates (d) similar to the gastral triradiates, only differing in the presence of apical rays. Apical ray short, curved upwards, sharply pointed, 60–90 μ long and about 7 μ thick at base.

Triradiates of oscular margin (e) strongly sagittal. Basal ray straight, slightly longer than paired rays, 180–240 μ long and 6–8 μ thick at base. Paired rays equal, widely divergent, 120–190 μ long and 6–8 μ thick at base.

Quadriradiates of oscular margin (f) like the triradiates of the same, but with a short apical ray, measuring 60–100 μ in length by about 7 μ thick.

Triradiates of the peduncle (g) strongly sagittal. Basal ray straight, longer than paired rays, 140–240 μ long and 6–8 μ thick at base. Paired rays nearly equal, either straight or slightly curved, 85–160 μ long and 6–8 μ thick at base.

Quadriradiates of the peduncle (h) exactly similar to triradiates of the same, differing only in the presence of apical rays. Apical ray short, curved upwards, 45–75 μ long and 6–8 μ thick at base.

Oxea at the distal end of flagellate chamber (i) elongate spindle-shaped, more or less curved, tapering to both ends, measuring 270–540 μ in length and 8–15 μ thick in the thickest parts.

Oxea of the peduncle (j) similar to those at the distal end of flagellate chamber. They measure 180–430 μ in length and are 6–13 μ thick in the middle parts.

Remarks:— This species appears to be closely related to *Sycon petiolutum* (HAECKEL)¹⁾ and to *S. pedicellatum* KIRK²⁾, but it may be distinguished from the first species by its external appearance and by the

¹⁾ *Sycandra ampulla* var. *petiolata* HAECKEL (1872), pp. 308–311, Taf. 52, figs. 2a–2t, Taf. 58, fig. 6.

²⁾ *Sycon pedicellatum* KIRK (1897), pp. 313–314, Pl. XXXI, figs. 1a, 1b; Pl. XXXII, figs. 1a–1c.

difference in the arrangement of the spicules in the peduncle. From KIRK's species, the present species may be easily distinguished by the absence of spongorhiza and by the difference of the shapes of the sub-gastral triradiates and of the oxea.

This species is named after Dr. URAGAMI, the collector of the type specimen.

Locality: — Matsushima Bay.

6. *Leucandra tomentosa*, n. sp.

(Pl. VIII, fig. 6, Textfig. 4)

In the collection there exist numerous specimens of this new species which were collected by professor HOZAWA in 1938 and by the writer in 1939 from the littoral zone of Mahanashi-jima. They are all of a closely similar appearance, though they vary from 5 mm. to 20 mm. in length. Each of them represents a solitary person of a spherical or oval form, showing at the upper end an osculum surrounded by a well-developed collar. The outer surface is very strongly hispid owing to the presence of long oxea projecting from it. The gastral surface which lines the rather narrow gastral cavity is smooth and is perforated by several large circular apertures of exhalant canals, measuring up to 1 mm. across. The sponge was covered with mud and thus the colour is grey when preserved in alcohol.

The most perfect specimen (Pl. VIII, fig. 6) which I make the type of the species represents a spherical form, with a height of about 15 mm. and a diameter of 14 mm. The osculum is surrounded by a fairly well-developed collar with a height of about 4 mm. The gastral cavity is irregular in shape and is rather narrow. The sponge wall is about 5 mm. in the thickest parts.

The following description is based upon the type specimen.

Structure: The canal system is leuconoid. The flagellate chambers are spherical with a diameter of 60-100 μ and are set thickly in the chamber layer.

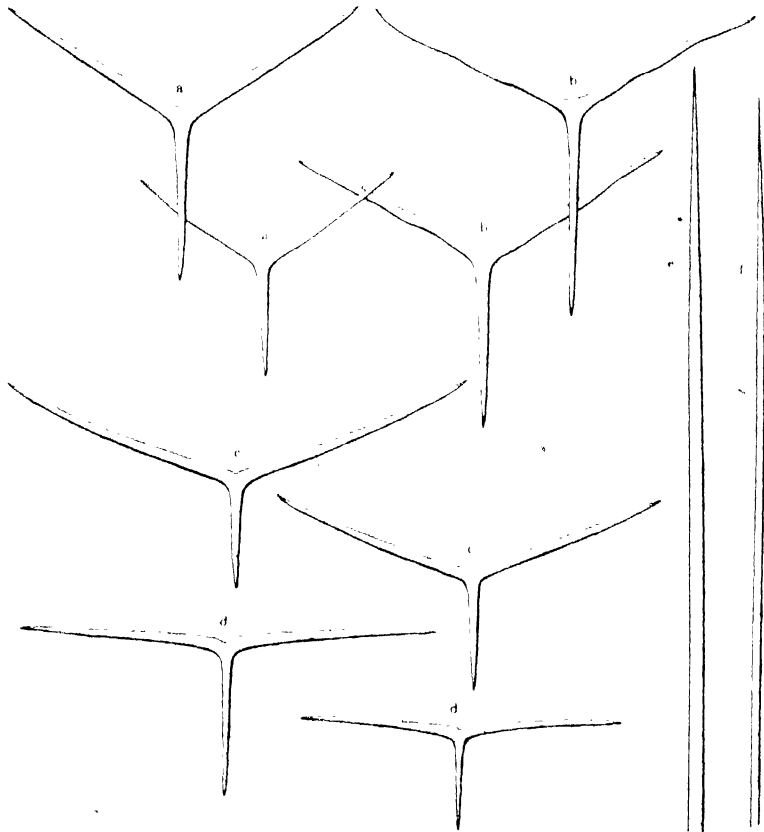
The dermal cortex is well defined due to the well-developed inhalant canals. The dermal skeleton is composed of tangentially arranged triradiates, large oxea and hair-like spicules. The outer portions of the latter two kinds of spicules project freely beyond the dermal cortex.

The skeleton of chamber layer is made up of triradiates only. They are densely and confusely packed in the layer.

The gastral skeleton is consisted of sagittal triradiates, which are closely set in several layers parallel to the gastral surface. The larger exhalant canals are lined with similar triradiates to those of the gastral surface.

The skeleton of oscular margin is composed of triradiates and linear spicules. The former are placed fairly densely with strongly divergent paired rays and with downwardly directed basal rays, while the latter spicules run parallel with the basal rays of the oscular triradiates and form a dense fringe.

Spicules (Textfig. 4): Dermal triradiates (a) slightly sagittal. All rays are nearly equally thick. Basal ray straight, shorter than paired



Textfig. 4. *Leucandra tomentosa*, n. sp. a, dermal triradiates; b, tubar triradiates; c, gastral triradiates; d, triradiates of oscular margin; e, Oxea of dermal surface; f, oxea of oscular margin. (a-d $\times 120$; e, f $\times 60$)

rays, 150–240 μ long and 8–12 μ thick at base. Paired rays equal, straight or slightly curved, 190–270 μ long and 8–12 μ thick at base.

Tubar triradiates (b) slightly sagittal. Basal ray straight, sharply pointed, either nearly equal to or slightly longer than paired rays, 200–270 μ long and 8–14 μ thick at base. Paired rays equal, more or less undulating, 190–250 μ long and 8–14 μ thick at base.

Triradiates of the larger exhalant canals exactly similar to those of the gastral cortex which will be mentioned later on.

Gastral triradiates (c) strongly sagittal. All rays are nearly equal in thickness and are lying in one plane. Basal ray straight, shorter than paired rays, 80–140 μ long and about 10 μ thick at base. Paired rays equal, widely diverging and are slightly curved forwards, 150–280 μ long and 10 μ thick at base.

Triradiates of oscular margin (d) sagittal. Basal ray straight, sharply pointed, shorter and thinner than paired rays, 100–240 μ long and 8–10 μ thick at base. Paired rays equal, strongly divergent, 200–380 μ long and 10–14 μ thick at base.

Large oxea projecting from dermal surface (e) straight, nearly uniformly thick in the greater parts of their length, tapering at the ends, measured 5 mm. long or more and 25–30 μ thick.

Hair-like oxea nearly straight, uniformly thick with both ends sharply pointed, about 5 mm. long and 2 μ thick.

Oxea of the oscular margin (f) nearly like as the large oxea of the dermal surface, differing only in size. They are about 3.5 mm. long and 12–17 μ thick in the middle parts.

Locality: — Mahanashi-jima in Matsushima Bay.

LITERATURE CITED

- DENDY, A. (1891). Monograph of Victorian Sponges. Part I. The Organization and Classification of the Calcareous Homocoela, with Descriptions of the Victorian Species. Trans. Roy. Soc. Vict., Vol. III, No. I, pp. 1–82, Pl. 1–11.
- HAECKEL, E. (1872). Die Kalkschwämme, eine Monographie. Berlin.
- HOZAWA, S. (1928). Report of the Biological Survey of Mutsu Bay. 6. Calcareous of Mutsu Bay. Sci. Rep. Tohoku Imp. Univ., Biol., Vol. III, No. 2, pp. 219–222, Pl. I.
- (1929). Studies on the Calcareous Sponges of Japan. Journ. Fac. Sci. Imp. Univ. of Tokyo, Sect. IV, Zoology, Vol. I, Part 5.
- (1940). On Some Japanese Calcareous Sponges. Sci. Rep. Tohoku Imp. Univ., Biol. Vol. XV, pp. 29–58, Pls. IV, V.
- KIRK, H. B. (1893). Contribution to a Knowledge of the New Zealand Sponges. Trans. New Zealand Instit., Vol. XXVI, pp. 175–179, Pl. XXII.

— (1897). Notes on New Zealand Sponges. Fourth Paper. Trans. New Zealand Instit., Vol. XXX, pp. 313-316, Pls. XXXI, XXXII.

EXPLANATION OF PLATE VIII.

- Fig. 1. *Leucosolenia mutsu* HOZAWA, $\times 2$.
Fig. 2. *Leucosolenia tehera*, n. sp. Natural size.
Fig. 3. *Sycon okadai* HOZAWA, $\times 1.5$.
Fig. 4. *Sycon matsushimense*, n. sp. $\times 2$.
Fig. 5. *Sycon uragamii*, n. sp. $\times 2$.
Fig. 6. *Leucandra tomentosa*, n. sp. $\times 1.5$.

1



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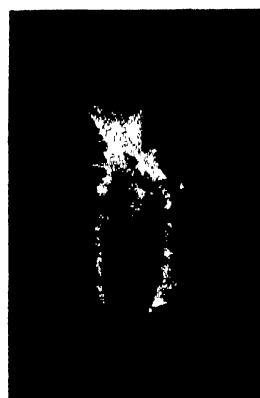
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TANITA photo.

S. TANITA: Calcareous of Matsushima Bay.

THE GENERAL CONSIDERATION OF THE DIURNAL ACTIVITY
OF THE STRAWBERRY WEEVIL, *ANTHONOMUS BISIGNIFER*
SCHENKLING, WITH SPECIAL REFERENCE TO THE BODY
TEMPERATURE OF THE WEEVIL AND THE ENVIRON-
MENTAL TEMPERATURE FACTORS

(DIURNAL RHYTHM OF ACTIVITIES IN INSECTS AND ITS
ENVIRONMENTAL CONDITIONS NO. IX)

By

MUTSUO KATÓ

Biological Institute, Tôhoku Imperial University, Sendai, Japan

(With 2 text-figures)

(Received February 2, 1940)

INTRODUCTION

Some kinds of insects are active in the daytime, while others are in the night. It seems to be generally understood that the periodicity found in the activity of insects correlates closely with the environmental conditions. It may be naturally thought that in order to investigate the said correlation we must first pay attention to the existence of or absence of sunshine. There are many investigations carried out about the relation between the light and the activity of the nocturnal insects. We have learned that a close correlation exists between this activity and the light intensity or the change of the same, and have also known the mechanism which causes the nocturnal activity by change of the light surroundings (YAGI, 1935, 1938).

It seems, however, to remain still doubtful whether the activity of the diurnal insects depends upon their light reaction or not. Most of the grasshoppers are generally considered to be the diurnal insects. According to ROCKWOOD (1925, cited from PARKER, 1930) a kind of grasshoppers is said to fly in unusually warm nights when the air temperature is approximately 26.7°C. This observation seems to suggest that the temperature may affect the diurnal activity of grasshopper. It is important to notice that the migration of grasshoppers is stopped and is again started when the passing clouds make the sun obscure and again exposed it, thus it was first thought to be caused by a light reaction, but it seems to be explainable rather by a temperature reaction when the air temperature

or the soil surface temperature is taken into consideration (PARKER, 1930). It was ascertained by BODENHEIMER (1930) that the body temperature of the grasshopper is on a fine morning fairly high compared with the air temperature. This seems to offer the important key to solve the problem above mentioned.

It must be very interesting and also important to investigate how the sunshine influences the activity of the diurnal insects, that is, whether the sunshine is effective as a light environment or as a heat environment.

For several years I have been studying the diurnal rhythm of activity seen in the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING, and have already published several papers concerning it. In the present paper I should like to deal with the general consideration of the diurnal activity of the Strawberry Weevil.

Before proceeding further I wish to express my sincere thanks to Prof. Dr. SANJI HÔZAWA for the kind instruction which I received from him in the course of the present investigation. I desire also to express my hearty thanks to Assist. Prof. Dr. ISAO MOTOMURA for his valuable criticisms.

THE DIURNAL RHYTHM OF ACTIVITIES IN THE WEEVIL AND THE METEOROLOGICAL CONDITIONS

1. *The Diurnal Rhythm of Activity*

The Strawberry Weevil is generally inactive during the night resting near the root of the strawberry plant or inside of the bract of flower-buds of the same. But after the sunrise it becomes suddenly active and begins to crawl and fly and also to lay eggs injuring the flower-buds. Towards the evening the activity becomes gradually weaker and then it goes into a resting condition for the night.

The egg-laying activity of this weevil is considered to be an index of its general activity. According to the statistical investigation (KATÔ, 1937a), the number of eggs laid during the night is only 11.6 percent of the total number of one day, and 88.4 percent of the same are laid during the daytime. The number of eggs laid in the morning (6th-10th hour) is 22.6 ± 1.2 percent of the total number of one day and the same in the midday (10th-14th hour) increases remarkably to 37.1 ± 1.7 percent, but the same in the evening (14th-18th hour) decreases to 28.7 ± 1.8 percent. Thus it must be understood that the difference between 22.6 percent of the morning and 28.7 percent of the evening is statistically significant.

In fine the Strawberry Weevil is evidently a diurnal insect and the diurnal rhythm of activities is clearly to be seen.

II. *Correlation Between the Activity and the Environmental Meteorological Conditions*

i. Duration of the sunshine.

By the first investigation (KATÔ, 1936) it was recognized that the duration of sunshine is the most important factor correlating with the egg-laying activity.

Then the duration of sunshine must be inquired into qualitatively. Namely, does the duration of the sunshine act upon the egg-laying activity as a light stimulus or as a heat stimulus? That is, is the diurnal rhythm of the activity governed by the light reaction or by the temperature reaction?

It is easily noticeable that in the case of this kind of weevil the eggs are laid at midnight on warm nights (KATÔ, 1938b). It is therefore permissible to think that the egg-laying may take place without any stimulation of light. This fact was also exactly noticed in the laboratory experiment (KATÔ, 1938b). If the temperature is kept fairly high, many flower-buds injured by the oviposition of the weevils will be found of strawberry plants which were placed in a dark room and also some crawling traces will be found on the smoked paper set for the experiment.

From the above facts it may be understood that the light is not very important for the egg-laying activity of the Strawberry Weevil, and consequently the heat energy must be taken up as the qualitative factor of the sunshine.

ii. The Solar Radiant Energy.

In the next experiment the solar radiant heat, the air temperature, and the soil surface temperature were taken into consideration. From the statistical investigation made on the correlation between the activity of the insect and the meteorological conditions in the course of one day (KATÔ, 1937a), we knew that the solar energy is the most effective upon the egg-laying activity.

During the time extending from 6th to 10th hour in the morning, the fluctuation of the amount of the solar energy is the most important factor controlling the activity, but in the noon-time covering from 10th to 14th hour the air temperature and the soil surface temperature, which

were not so notable in the morning, act rather effectively upon the said activity together with the solar energy. Then in the evening the air temperature and the soil surface temperature become the main controlling factors.

We know finally that the solar radiant heat is most effective in the period extending from 6th to 14th hour and the air temperature and the soil surface temperature are effective mainly in the period covering from 10th to 18th hour (Table 1).

TABLE 1.

Coefficients of correlation calculated between the climatic factors and the activity of the oriposition of the Strawberry Weevil.

Time-interval	Solar radiant energy	Soil surface temperature	Air temperature	Evaporation	Humidity
h h					
6.00 10.00	0.884 ±0.032	0.645±0.084	0.822±0.047	0.928±0.020	-0.631±0.087
10.00 14.00	0.913 ±0.023	0.932 ±0.018	0.915 ±0.023	0.921±0.023	-0.766±0.058
14.00 18.00	0.740±0.061	0.839 ±0.042	0.825 ±0.045	0.719±0.068	-0.561±0.096

(adapted from KATO, 1937a)

It seems to be permissible to think that the air temperature and the soil surface temperature are fairly low in the morning and do not accelerate the activity of the weevils and, thus, the solar radiation only acts as the temperature factor upon the said activity. But with the progression of time, the air temperature and the soil surface temperature rise gradually and attain the temperature zone of activity of the weevil, and then the influence of the solar radiation is not so strong as in the morning. This may be the reason why the air temperature and the soil surface temperature increase in correlation with the said activity in the noon-time and in the evening.

iii. The Locomotion Velocity and the Solar Radiation.

Now we must pay attention to the fact that the solar energy influences the activity in an added condition to the environmental temperature expressed mainly by the air temperature or by the soil surface temperature. The activity expressed in the sunshine soon falls in potency in the temperature environment expressed by the air temperature, if the sunshine is intercepted. On the contrary, the activity of the same in the temperature environment without sunshine, rises quickly and remarkably

when the sun begins to shine. This is obviously observed in the experiment carried out on the locomotion velocity of the weevil (KATÔ, 1938b). We learned from the above experiment that the locomotion velocity correlates closely with the presence or absence of the sunshine or with the reading of the black heliothermometer, and furthermore that the locomotion velocity is exactly swayed by the air temperature when the sun is obscured temporarily (Fig. 1).

iv. The Egg-laying Activity in the Case of the Solar Eclipse.

In the case of the solar eclipse it is observed that the diurnal rhythm of the meteorological condition is divided into two parts by the decreasing of the solar radiation caused by the eclipse. In order to clarify the correlation between the activity and the environmental conditions, it seems to be of interest to investigate whether the diurnal rhythm of the activity of the weevil is affected or not by the said meteorological conditions caused by the solar eclipse.

According to the experiments executed in the case of a total eclipse which occurred on the 19th of June 1936 in some parts of Hokkaido (KATÔ, 1937b), it was known that the diurnal rhythm of the egg-laying activity of the Strawberry Weevil was exactly divided into two parts being affected by the changes of the temperature factors, especially by the decreasing of the solar radiation, and it was also known that the periodicity of the said activity was governed only by the diurnal variation of the meteorological conditions, mainly by the temperature factors.

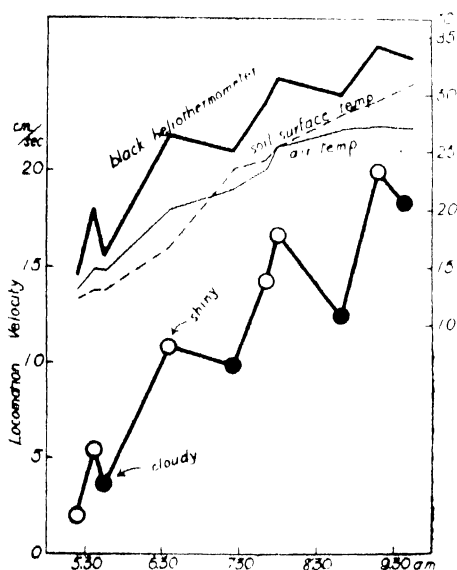


Fig. 1. Showing the correlation between the locomotion velocity of the weevil and the temperature environments. It is seen that the velocity under the cloudy weather rises in accordance with the rise of the air temperature. adapted from KATÔ, 1938b

v. The Transformation from the Nocturnal Resting Condition to the Diurnal Active Condition.

From the investigation carried out on the process of transformation from the nocturnal resting condition to the diurnal active condition (KATÔ, 1938b), it was known that the said transformation progresses in cloudy weather uniformly and gradually depending upon the gradual rising of the air temperature or of the soil surface temperature, but in spite of the low air temperature, the resting condition changes on a fine morning irregularly and quickly being influenced by the solar radiation. It was also observed that the transformation occurring in the morning is generally quick, but in the evening the active condition changes gradually into an inactive one, even if the solar energy weakend quickly. This seems to depend upon the fairly high air temperature and also to be caused by the slow falling of the same.

It may be concluded from the results above mentioned that the activity of the weevil is surely governed by the temperature reaction of the weevil itself and also that the solar radiation is very effective upon the said activity. We learn also that a fairly strong absorption power of the solar radiation is seen in the case of the weevil.

THE BODY TEMPERATURE OF THE WEEVIL AND THE ENVIRONMENTAL CONDITIONS

1. *Body Temperature of the Weevil.*

It was clarified from the above experiments that the diurnal variation of the activity of the weevil is caused by the temperature reaction and thus the diurnal rhythm of the activity coincides with the diurnal variation of the environmental temperature factors. It may be also conceivable that the solar radiant energy which might have been neglected is rather important as one of the temperature factors. Consequently it may be permissible to think that the solar radiant energy must be taken up as an environmental temperature factor in the field of ecological investigation.

From the view point above mentioned the ecological investigation on the body temperature of the weevil becomes naturally necessary.

It was concluded from the preliminary investigation (KATÔ, 1939) that the change of body temperature is caused primarily by the increase and decrease of the solar radiant energy, the other environmental factors acting only secondarily. It may be needless to say that in this case the

air temperature forms the base of the body temperature and the solar energy is effective in an added condition to the air temperature.

If the activity of the weevil is exactly due to the temperature reaction, the body temperature estimated from the experimental results concerned with the correlation between the activity and the environmental temperature factors may certainly agree with the body temperature measured directly.

The locomotion velocity was measured in the laboratory and also in field conditions (KATÔ, 1938b) (Fig. 2). When we compare the similar velocity obtained in the laboratory work and in the field experiment it is known that the air temperature in the case of field condition is fairly low compared with that in the laboratory experiment. If the velocity is equal in each case, it may be permissible to think that the body temperature must be equal in both cases.

Now in the case of the laboratory work it was recognized that the air temperature represents the body temperature itself. Accordingly in the case above mentioned the body temperature is surely higher in the field condition than the environmental air temperature. In Fig. 2 the horizontal distance between **A**-group and **B**-group shows the difference between the air temperature and the body temperature. It is known from this that the body temperature is 3°C. or some what higher than the air temperature, and also that this is in accord with the direct measurement of the body temperature (KATÔ, 1939, 1940).

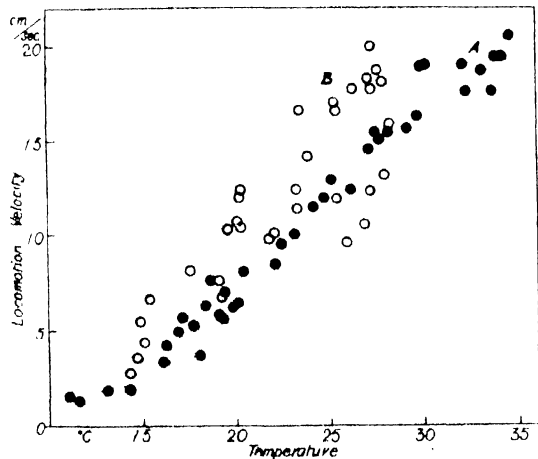


Fig. 2. Showing the locomotion velocity of the weevil measured under field conditions and in the laboratory. **A**, the results obtained by the laboratory work under the gradually rising temperature. **B**, Relation between the velocity measured in the field and its environmental air temperature. (adapted from KATÔ, 1938b)

II. *Diurnal Rhythm of the Body Temperature and the Environmental Temperature Factors.*

A parallelism was shown to exist between the fluctuation of the body temperature and the variation of the temperature factors, especially of the solar energy, and it was also recognized that the environmental temperature is effective not directly but indirectly through the body temperature upon the activity.

But we are still not thoroughly convinced of the existence of a correlation between the activity of the weevil and the environmental conditions without having the experimental result of the diurnal variation of the body temperature.

From the investigation made concerning the diurnal variation of the body temperature (KATÔ, 1940) it was concluded that, regarding the environmental temperature factors which controll the body temperature, the solar radiant energy is predominant in the morning, but with the progression of time the air temperature becomes gradually important and finally it becomes a prevailing factor in the evening.

In spite of a remarkable utilization of the solar energy, the body temperature in the morning is lower than in the evening, as the air temperature, which forms the base of the body temperature, is fairly lower in the former than in the latter.

It is also known that the body temperature rises rapidly in the morning being influenced by the solar radiation, but in the evening it falls rather slowly depending upon the gradual falling of the air temperature.

RELATION BETWEEN THE ACTIVITY OF THE WEEVIL AND THE BODY TEMPERATURE OF THE SAME

We have already learned that a correlation exists between the activity and the environmental factors and also between the body temperature and the environmental conditions. Now the relation between the activity and the body temperature of the weevil must be fully considered.

From the statistical investigation made concerning the correlation between the egg-laying activity and the environmental meteorological factors, it was known that in the morning the air temperature or the soil surface temperature is not so effective but the solar energy is the prevailing factor controlling the activity, and that at about noon the former becomes equally as important as the latter, and this tendency grows conspicuous in the evening, that is, both the air temperature and the soil

surface temperature become the predominant factors controlling the egg-laying activity (KATÓ, 1937a) (Table 1). The fact above mentioned may be very comprehensive by the fact that the environmental temperature factors act upon the activity of the weevil through the body temperature, and, namely, by the study of the diurnal variation of the body temperature of its controlling temperature factors.

In the morning the air temperature or the soil surface temperature, which forms the base of the body temperature, is fairly low, and thus a remarkable utilization of the solar radiation may be observed. Thus the solar radiant heat influences strongly the body temperature and consequently the activity through the body temperature. But with the progression of time the air temperature rises gradually and then the body shows a fairly high temperature even if the sunshine is absent. Furthermore a kind of temperature regulation is seen inhibiting the absorption of the solar radiation caused by the high temperature. In the evening a remarkable utilization of the solar radiant heat is not seen, as the air temperature is still fairly high and the body temperature is also high even though there is no sunshine. Thus the air temperature becomes important with the progression of time as one of the controlling factors of the body temperature and acting strongly upon the activity of the weevil through its body temperature.

It was already mentioned that the resting condition of the night is changed rapidly into the active condition of the daytime in the case of the fine weather, but it is done rather gradually in the case of a cloudy morning. This is also explainable by the rapid or slow velocity of the rising of the body temperature. That is to say in the case of a fine morning the body temperature rises rapidly utilizing strongly the solar radiation, and consequently the activity of the weevil increases quickly, but on the contrary in cloudy weather the available solar energy is very weak and the body temperature rises gradually with the rise of the air temperature and thus the transformation of the activity is very slow.

It is generally seen that the diurnal active condition changes very slowly into the nocturnal inactive condition. This may be explained by the slow falling of the air temperature and consequently by the gradual fall of the body temperature.

Regarding the egg-laying activity, the number of flower-buds injured by oviposition is greater in the evening than that in the morning. Although the amount of the solar energy is almost equal in both periods, the air temperature which forms the base of the body temperature is

fairly high in the evening compared with that in the morning. Accordingly the body temperature itself is lower in the morning than in the evening in spite of the remarkable utilization of the solar radiation, and consequently the egg-laying activity may be more active in the evening than in the morning.

CONCLUSION

The diurnal activity shown by the Strawberry Weevil is due to the temperature reaction of the weevil. It may be therefore recognized that the temperature factors of the meteorological environments act strongly upon the activity of the said weevil.

As these temperature factors are affective upon the activity through the body temperature of the weevil, the diurnal activity progresses in accordance with the diurnal variation of the body temperature.

It may be conclusively permissible to think that, in the case of the insects whose activity depends upon their temperature reaction, the environmental temperature factors influence externally upon the said activity and the body temperature is internally effective upon it.

It is also noted that the solar radiant energy which has been almost neglected in the field of ecology must be taken into consideration as one of the environmental temperature factors together with the air temperature or the soil surface temperature.

LITERATURE CITED

- BODENHEIMER, F. S. (1930). Studien zur Epidemiologie, Oekologie und Physiologie der afrikanischen Wanderheuschrecke (*Schistocerca gregaria* FORSK.). Zeitschr. angew. Entom., Bd. 15, 1-123.
- KATÔ, M. (1936). On the Activity of Oviposition of the Strawberry Weevil, *Anthonomus bisignatus* ROELOFS (Diurnal Rhythm of Activities in Insects and its Environmental Conditions No. 1). Sci. Rep. Tôhoku Imp. Univ., Biol., Vol. X, 697-708.
- , (1937a). A Statistical Investigation of the Correlation between Climatic Conditions and the Egg-Laying Activity of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING (" No. 2). Ibid., Vol. XI, 307-321.
- , (1937b). Changes Occurring in the Egg-Laying Activity of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING, in the Case of a Solar Eclipse (" No. 3). Ibid., Vol. XI, 353-359.
- , (1938a). The Temperature Limit of Activity of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING (" No. 4). Ibid., Vol. XII, 501-510.
- , (1938b). The Diurnal Activity of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING, with a Note of the Ecological Meaning of the Solar Radiant Energy (" No. 5). Ibid, Vol. XII, 511-530.

- , (1939). Body Temperature of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING, and its Limiting Factors (" No. 6). Ibid., Vol. XIV, 11-19.
- , (1940). Diurnal Variation in the Body Temperature of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING (" No. 8). Ibid., Vol. XV, 97-103.
- PARKER, J. R. (1930). Some Effects of Temperature and Moisture upon *Melanoplus mexicanus mexicanus* SAUSSURE and *Camnula pellucida* SCUDDER (Orthoptera). Univ. Montana Agric. Exp. Stat., Bull., No. 223.
- YAGI, N. (1935). On the Nocturnal Activity of Moth of *Chilo simplex* BUTLER. Journ. Imp. Agric. Exper. Stat., Vol. II, 471-490.
- , (1938). On the Structure and Dioptries of Compound Eye of *Chilo simplex* BUTLER. Ibid., Vol. III, 183-206.

FEEDING ACTIVITY OF A GRASSHOPPER, *PRUMNA* SP., WIDELY DISTRIBUTED AT MTS. HAKKÔDA*

(DIURNAL RHYTHM OF ACTIVITIES IN INSECTS AND ITS
ENVIRONMENTAL CONDITIONS NO. X)

By

MUTSUO KATÔ

Biological Institute of the Tôhoku Imperial University, Sendai

(With 8 text-figures)

(Received February 10, 1940)

INTRODUCTION

It seems to be generally accepted that the activity of grasshoppers is remarkably influenced by the temperature environments. Namely the said activity is considered to be governed by the temperature reaction of the grasshopper itself. In this case we must pay attention to the fact that the environmental temperature is not effective directly upon the activity but through the body temperature. That is to say we are not able to understand thoroughly the act of the environmental temperature without taking the body temperature of the insect into consideration.

An investigation of the temperature limit of various stages seen in the activity of some Locusts was carried out by HUSSEIN (1937) and it seems to suggest the reliability of the opinion being mentioned by UVAROV who has expressed his opinion in the preface of the paper which dealt with the above investigation. From the ecological investigation made about the diurnal activity of the Strawberry Weevil it was concluded that it is very important to inquire into the reciprocal relation of the insect activity, the body temperature of the insect and the environmental temperature factors, in order to know the insect activity which is controlled mainly by the temperature reaction (KATÔ, 1940).

FEEDING ACTIVITY OF GRASSHOPPERS

According to PARKER (1930) the temperature zone of feeding activity is in accord with that of the general activity. The Lessor Migratory Locust begins to feed when the air temperature and the soil surface tem-

*Contribution from the Mt. Hakkôda Botanical Laboratory, No. 28.

perature become respectively from 12.7 to 17.2°C., and from 21 to 34.4°C. The temperature ranging from 20 to 25.5°C. and from 40 to 44.4°C. were found to be favorable for the maximum feeding. But the feeding activity decreases rapidly when the temperature becomes higher. In these cases, however, the air temperature was measured at the height of 1 feet above the ground and thus it does not show the temperature at the height of 2 inches above the soil surface where the grasshoppers are found most abundantly.

A kind of grasshopper, *Prumna* sp.¹, is widely distributed at Mts. Hakkôda, Aomori Prefecture, North-East Honshû, being found within the area extending from a level about 700 metres above the sea to the summit of ÔDAKE, the main mountain of Mts. Hakkôda, and 1,585 metres

TABLE 1.

The specific names of plants which may be injured by Prumna sp.

<i>Salix Reinit</i>	Salicaceae
<i>Polygonum Weyrichii</i>	Polygonaceae
<i>Glaucidium palmatum</i>	Ranunculaceae
<i>Hydrangea paniculata</i> var. <i>floribunda</i>	Saxifragaceae
<i>H. macrophylla serrata</i> f. <i>acuminata</i>	"
<i>Sorbus Aucuparia</i>	Rosaceae
<i>Daphniphyllum humile</i>	Euphorbiaceae
<i>Ilex leucolada</i>	Aquifoliaceae
<i>I. Sugeroki brevipedunculata</i>	"
<i>Acer japonicum</i> var. <i>typicum</i>	Aceraceae
<i>A. Tschonoskii</i>	"
<i>Viola brevistipulata</i>	Violaceae
<i>Acanthopanax sciadophyllodes</i>	Araliaceae
<i>Menziesia ciliicalyx</i> var. <i>multiflora</i>	Ericaceae
<i>Vaccinium Smallii</i>	"
<i>V. axillare</i>	"
<i>Rhododendron Albrechtii</i>	"
<i>R. Fauriae</i> var. <i>rufescens</i>	"
<i>Leucothoe Grayana</i> var. <i>typica</i>	"
<i>Tripetaleia bracteata</i>	"
<i>Fauria Crista-galli</i>	Gentianaceae
<i>Menyanthes trifoliata</i>	"
<i>Viburnum furcatum</i>	Caprifoliaceae
<i>Petasites gigantus</i>	Compositae
<i>Cacalia adenostyloides</i>	"
<i>Aster Glehni</i>	"
<i>Arnica unalaschensis</i>	"
<i>Lysichiton camtschatense</i>	Araceae
<i>Arisaema heterophyllum</i> var. <i>typicum</i>	"
<i>Aruncus silvestor</i> var. <i>americanus</i>	"
<i>Hosta japonica</i> var. <i>angustifolia</i>	Liliaceae

¹It was ascertained from Mr. H. FURUKAWA that this grasshopper will be reported by himself in the near future as a new species under the specific name of *Prumna uzume* (Japanese name: Towada-Maruo-Huki-Batta).

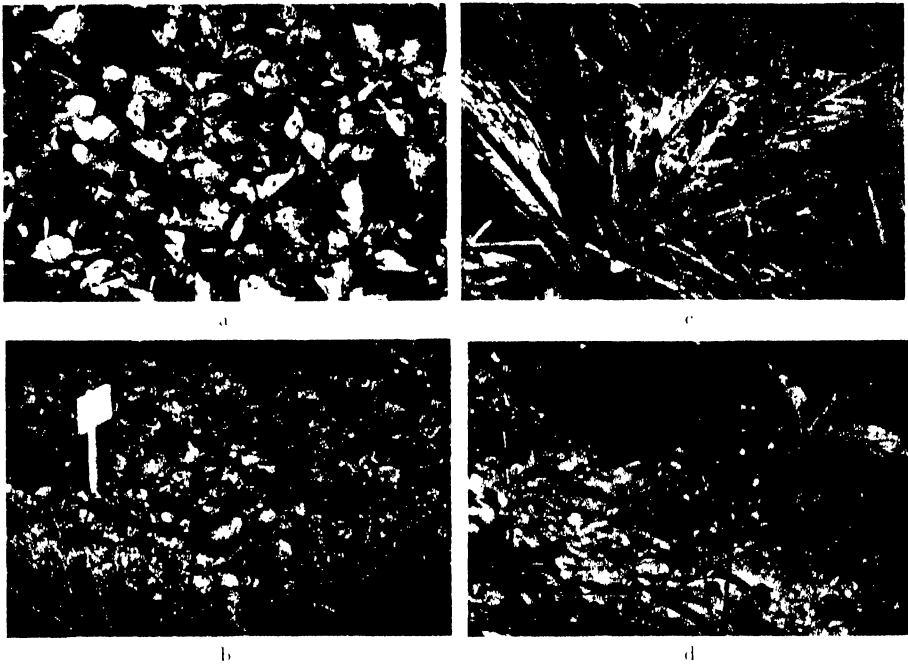


Fig. 1 Showing some kinds of plants injured by *Prunna* sp. a, *Viola brevistipulata* W. BECK. b, *Fauria Crista-galli* MAKINO, c, *Lysichiton camtschatense* SCHOTT d, *Hydrangea paniculata* var. *floribunda* REGEL.

high above the sea. The density of population of this grasshopper is thickest at Sukayu Hot-Spring or around the Botanical Garden of the Tôhoku Imperial University situated in the level of from about 900 metres to 950 metres above that of the sea.

This grasshopper seems to be very polyphagous and the number of plants which may be injured by it run up to 31 species according to the observation done in the said botanical garden (Table 1; Fig. 1 & 2).

I have wished before to investigate the relation between the feeding activity of this grasshopper and the meteorological conditions, in other words to know whether the feeding



Fig. 2 Some leaves of *Hydrangea* showing the typical injury made by *Prunna* sp.

activity of this grasshopper is governed mainly by the environmental temperature factors, or by the temperature reaction of the grasshopper itself.

I was able to have an opportunity to carry out this investigation by the kindness of Prof. Dr. YOSHII YOSHII, the director of the Mt. Hakkôda Botanical Laboratory, and I am very grateful to him. I wish here to acknowledge the kind instruction and encouragement given by Prof. Dr. SANJI HÔZAWA and Assist. Prof. Dr. ISAO MOTOMURA. I am also grateful to Mr. HARUO FURUKAWA for his kind information about the specific name of this grasshopper.

METHOD AND MATERIAL

The rearing equipment is rather simple consisting of a porous pot and a bell-shaped cage made of wire. The pot is 25 cm. in diameter and is 12 cm. in height being filled with soil. The bell-shaped cage is 20 cm. in diameter and 30 cm. in height and was made to cover the pot (Fig. 3).



Fig. 3. Showing the apparatus used in the field experiment which was executed in the back yard of the Mt. Hakkôda Botanical Laboratory

Hidrangea paniculata var. *floribunda* REGEN was used as food for the experimental insects, as it was found to be most satisfactory in carrying out this investigation. A twig taken from this plant was put in water contained in a glass bottle which is 2 cm. in diameter and 7 cm. long and was preliminary settled in

the soil of the porous pot. Twenty individuals males and females were confined in each of the said rearing cages.

The experiments were executed in the latter part of August of 1939. In this season the grasshoppers seemed to be at their full growth.

The amount of food consumed by the insect was measured by a special method. A twig with some leaves which was to be given to the insect as food was first confined in a bottle saturated with moisture for about 24 hours and then it was measured in weight. The above was given to the insect for some definite duration of time. After that it was again

confined in the bottle above mentioned for the same period as before and its weight measured again. Thus the amount of food consumed in some definite time may be represented by the difference in the weights obtained in the two ways as mentioned above, in reality it means the weight of raw matter devoured by the insect.

Of the environmental conditions the temperature factors were mainly observed. The solar radiant heat was measured using a black heliothermometer and the air temperature was observed at a height of about 10 cm. above the ground. The soil surface temperature was also measured. These observation above mentioned were all made in the said rearing cage. The evaporation and wind velocity were also measured.

RESULTS AND DISCUSSION

1. Diurnal rhythm in the Feeding Activity.

The fluctuation of the feeding activity during the summer of 1939 extending from 15th of August to 25th of the same month was shown in Fig. 4. The feeding activity was represented by the amount of food consumed per four hours in the daytime, but the amount of food consumed during the night from 18th to 6th hour of the next morning was divided

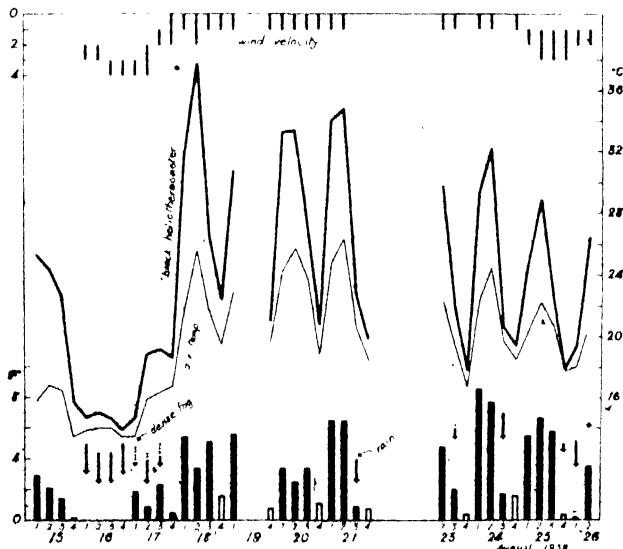


Fig. 4. Frequency histogram showing the amount of food consumed by *Prumna* sp. during the summer of 1939 and the meteorological conditions of the environments. 1: 6th-10th hour, 2: 10th-14th hour, 3: 14th-18th hour, 4: 18th-6th hour.

into three parts for the convenience of comparing it with that of the daytime.

There is not a doubt that the feeding is done mostly in the daytime. It is, however, observed that very often the feeding activity becomes rather weak in the midday. According to the data obtained from the field experiment the amount of food consumed in the daytime is 81.75 percent of the total of one day: being 33.56 percent in the morning extending from 6th to 10th hour, 29.90 percent in the noon-time extending from 10th to 14th hour and 21.29 percent in the evening covering from 14th to 18th hour. During the night it was only 15.24 percent of the total amount of food consumed in one day.

According to the control experiment executed in the laboratory, 70.08 percent of the total amount of food of one day was taken in the daytime, and in the night the amount of food was 29.19 percent, these are pretty large when compared with the figures obtained from the field experiment.

This difference may be understood when the temperature environment is considered. In the field the fluctuation of temperature is rather great and moreover the temperature falls markedly in the night. But in the room the fluctuation of temperature is small and the temperature in the night is rather high. Finally it may be permissible to think that the feeding activity of this kind of grasshopper is mainly governed by the temperature environment.

II. *Relation Between the Feeding Activity and the temperature environments.*

From Fig. 4 we can see the parallelism existing between the fluctuation of the feeding activity and the variation of the temperature environments except for the inverse phenomenon often observed in the midday. The rainfall may be looked upon as a factor inhibiting almost all of this activity.

Fig. 5_E shows the mean value of the feeding activity obtained from the field experiment, excluding the data obtained on a rainy day. Fig. 5_A shows the same secured in the laboratory experiment. Both of these figures show the diurnal variation of the amount of food consumed per four hours.

The type of these diurnal variations is obviously different from each other. In the case of the laboratory work the fluctuation of the feeding activity is exactly in accordance with the variation of the air temperature,

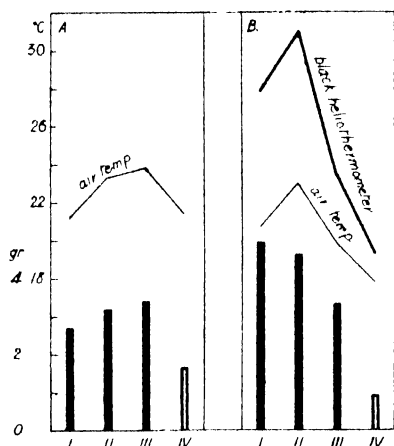


Fig. 5. Showing the diurnal variation of the feeding activity of *Prumna* sp. No. 1. A, laboratory work, B, field experiment; I: 6th-10th hour, II: 10th-14th hour, III: 14th-18th hour, IV: 18th-6th hour.

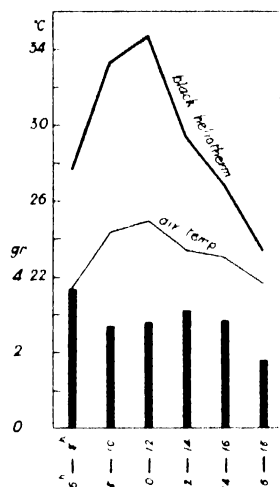


Fig. 6. Showing the variation of the feeding activity of *Prumna* sp. No. 2.

but in the case of the field condition this activity is not parallel with the process of the variation of temperature factors. Fig. 6 shows the diurnal variation of the feeding activity represented by the amount of food consumed per two hours, the data being obtained during the period from 18th to 20th day of August. Here may be found also the discordance in both diurnal variation of the feeding activity and of the temperature factors. The maximum feeding is seen twice a day, one in the morning and the other in the evening; and the activity is weakened at noon time. This phenomenon is seen clearly in the data obtained on 18th of August (Fig. 7). The amount of food consumed was largest in the period extending from 6th to 10th hour and also from 14th to 18th hour. In the period covering from 10th to 14th hour when the reading of the black heliothermometer went up to 37.7°C. and the

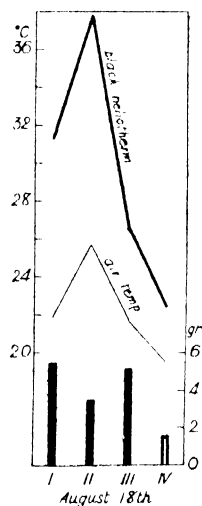


Fig. 7. Showing the relation between the feeding activity and the temperature environments. No. 1. The time interval is the same as that shown in Fig. 5.

air temperature was 25.7°C., the amount of food consumed decreased inversely to the high temperature.

If we compare in detail the diurnal variation of the feeding activity with that of the temperature environments, we may be able to know that the activity is surely inhibited by the high temperature during the noon-time. The feeding activity seems to decrease when the reading of the black heliothermometer goes up above 33–35°C. and the air temperature rises above 25–26°C.

Fig. 8 shows the results obtained on the 20th and 25th of August. Comparing these two data we make sure of the fact that the feeding

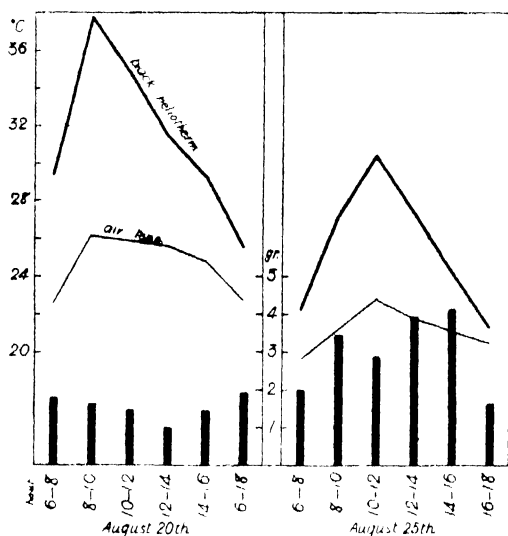


Fig. 8. Showing the relation between the feeding activity and the temperature environments. No. 2. The time interval is the same as that shown in Fig. 5.

was observed only in the noon-time (from 10th to 12th hour) and consequently the figure to show the diurnal variation of the feeding activity became the Δ -type.

In the laboratory experiment the temperature does not rise so high as to cause the decrease of the feeding activity, and thus the Δ -type variation of diurnal activity is seen in this case.

activity is governed by the temperature reaction and also that high temperature inhibits the feeding activity. On 20th of August, it was very fine, the sunshine was very bright, but the temperature environment was too high for the grasshopper, being out of the optimum temperature zone of feeding activity. Thus the diurnal variation of the activity represents the ∇ -type, showing the maximum amount of food in the period extending from 6th to 8th hour and from 16th to 18th hour. But on the 25th day of August, the inhibiting action caused by high temperature

III. *Solar Radiant Energy as a Environmental Temperature Factor.*

I have already mentioned in my previous papers that, in the ecological investigation of the insect in which the activity is governed by the temperature reaction of the insect itself, we must pay attention to the body temperature of the insect internally and to the temperature environments externally; and thence we learn that the solar radiant energy, which has been almost neglected in the field of ecology, becomes very important in acting as one of the temperature factors.

The relation between the environmental temperature and the amount of food consumed per four hours is tabulated in Table 2.

TABLE 2.

The relation between the environmental temperature and the amount of food consumed per four hours.

Laboratory experiment		Field experiment			
Air temp.	Amount of food	Air temp. (reading of black heliotherm.)	Amount of food	Reading of black heliotherm. (air temp.)	Amount of food
16-18°C.	- gr.	16-18°C. (20.1°C.)	1,140 gr.	16-18°C.	- gr.
18-20	-	18-20 (21.1)	1,398	18-20	-
20-22	1.945	20-22 (26.3)	5,040	20-22 (19.3°C.)	1,960
22-24	2.690	22-24 (29.3)	5,830	22-24 (18.7)	3,575
24-26	4.378	24-26 (34.1)	4,702	24-26 (17.7)	3,473
26-28	-	26-28 -	-	26-28 (21.9)	4,303
28-30	-	28-30 -	-	28-30 (22.4)	6,750
30-32	-	30-32 -	-	30-32 (23.1)	6,230
32-34	-	32-34 -	-	32-34 (25.6)	5,250

We can see that the amount of food consumed in the field experiment is greater than that consumed in the laboratory, when we observe both under similar air temperature. This may be understood by the fact that the activity is governed by the temperature reaction.

In the case of the laboratory experiment we may be satisfied if we take up only the air temperature as the environmental temperature factor, and it may be permissible to think that the air temperature shows the body temperature itself, as it does not fluctuate so much and also the fluctuation is carried very gradually.

In the field experiment, however, we must take up the air temperature, soil surface temperature and the solar radiant heat as the temperature factors, and in this case the body temperature rises fairly high com-

pared with the air temperature, being influenced by the solar radiant heat.

For instance in the case of the field experiment the amount of food consumed was about 5 gr. when the air temperature was 20-22°C. and was greater than that in the case of the room temperature 24-26°C. high. This shows that the body temperature is higher than 24-26°C. being influenced by the solar radiation which is 26.3°C. by the black heliothermometer. On the contrary, the amount of food consumed is about 4 gr. when the black heliothermometer is 26-28°C. and this is almost equal to the amount of food consumed in the case of the room temperature 24-26°C. high. From this fact we may assume that the body temperature may be about 24-26°C. and it is fairly low compared with the reading of the black heliothermometer. It is however needless to say that this body temperature is of course higher than the air temperature of about 22°C. high.

It must be noted that in the case of the field experiment the feeding activity is very weak when the air temperature is below 20°C. This phenomenon may depend upon the fact that such low temperature as 20°C. or below may be observed only on a cloudy day or during the night and thus the feeding activity is done under the temperature environment with very weak solar radiation or without it.

SUMMARY

In the present paper the relation between the feeding activity of a grasshopper, *Prumna* sp., and the meteorological environment was dealt with.

The feeding activity is mainly governed by the temperature reaction, and its diurnal variation represents during the summer time the **M**-type, which has two maximum, one in the morning and the other in the afternoon, and one minimum during the midday being caused by the high temperature.

LITERATURE CITED

- HUSSEIN, M. (1937). The effect of temperature on Locust activity (with a preface by UVAROV, B. P.). Ministry of Agric., Egypt, Technical and Scientific Service, Bull. No. 184, 55 pp.
- KATŌ, MUTSUO (1940). The general consideration on the diurnal activity of the Strawberry Weevil, *Anthonomus bisignifer* SCHENKLING, with special reference to the body temperature of the weevil and the environmental temperature factors (Diurnal rhythm of activities in insects and its environmental conditions No. 9). Sci. Rep. Tohoku Imp. Univ., Biol., Vol. XV, 179-189.
- PARKER, J. R. (1930). Some effects of temperature and moisture upon *Melanoplus mexicanus mexicanus* SAUSSURE and *Camnula pellucida* SCUDDER (Orthoptera). Univ. Montana Agric. Exp. Station, Bull. No. 223.

WATER MITES FROM IZU

I. RHEOPHILOUS WATER-MITES FROM RIVER INÔZAWA

By

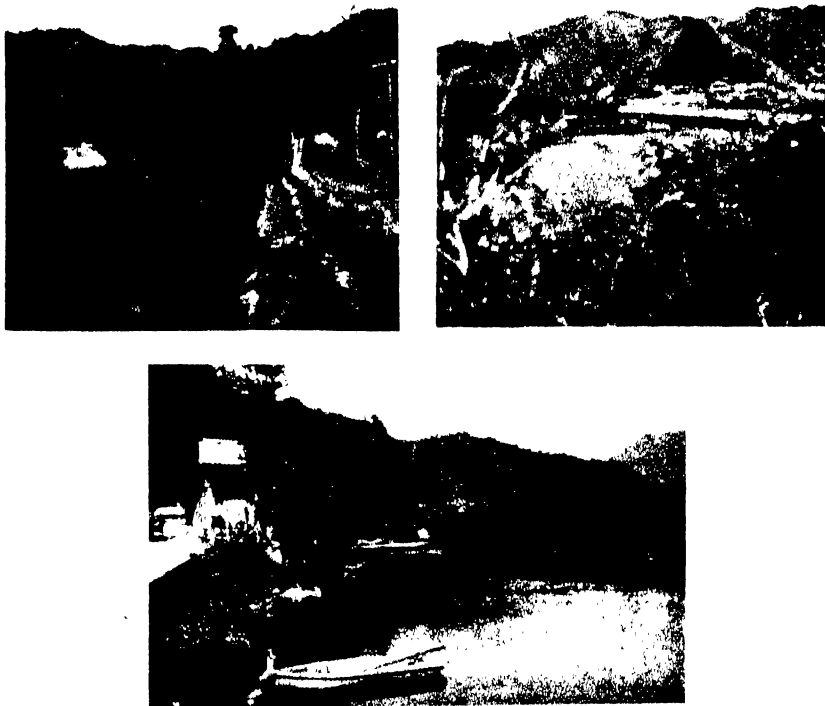
MASASI ENAMI

Mitsui Institute of Marine Biology, Simoda, Izu, Japan

With 16 text-figures

Received March 1, 1940

In the year 1939, a short interval occurring during the preparation of a physiological work, the writer had the pleasant experience of investigating a collection of water-mites obtained in the Izu Peninsula. The results will



Text-fig. 1. Three views of the main stream of the River Inôzawa, Izu.

appear separately in several successive papers, as the writer is unfortunately not at present able to devote the sufficient time necessary for the study of the whole number of specimens.

The present report on rheophilous water-mites is based chiefly upon materials collected by the writer himself on Jun. 8th, 1938 along the course of the River Inôzawa flowing into the Bay of Simoda. Of the twelve species, seven are introduced to the scientific world of this country for the first time, and three are likely to form new categories.

The species herein dealt with are listed as follows:

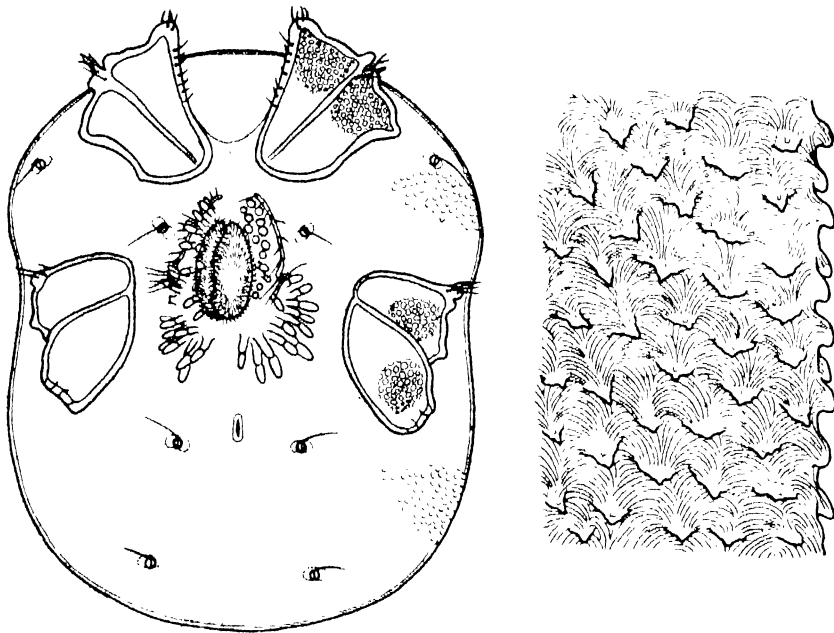
1. *Calonyx japonicus* (UCHIDA)
2. *Lebertia* (*Pilolebertia*) *leioderma* VIETS
3. *Atractides* (*Atractides*) *ellipticus* (MAGLIO)
4. *Atractides* (*Atractides*) *stadleri* WALTER
5. *Atractides* (*Atractides*) *nipponicus*, n. sp.
6. *Atractides* (*Rusetria*) *semisutus* SOKOLOV
7. *Hygrobates* (*Hygrobates*) *calliger* PIERSIG
8. *Megapus* (*Megapus*) *izuensis*, n. sp.
9. *Brachypoda versicolor* (MULLER) var.
10. *Aturus miyashitai* UCHIDA
11. *Aturus caudatus*, n. sp.
12. *Kongsbergia materna* THOR

To Dr. T. UCHIDA the writer is under deep obligation for valuable criticism and continuous encouragement. Also thanks are due to Dr. S. HÔZAWA who has shown high appreciation of, and interest in the writer's study of the water-mites.

1. *Calonyx japonicus* (UCHIDA)

(Text-figs. 2-5)

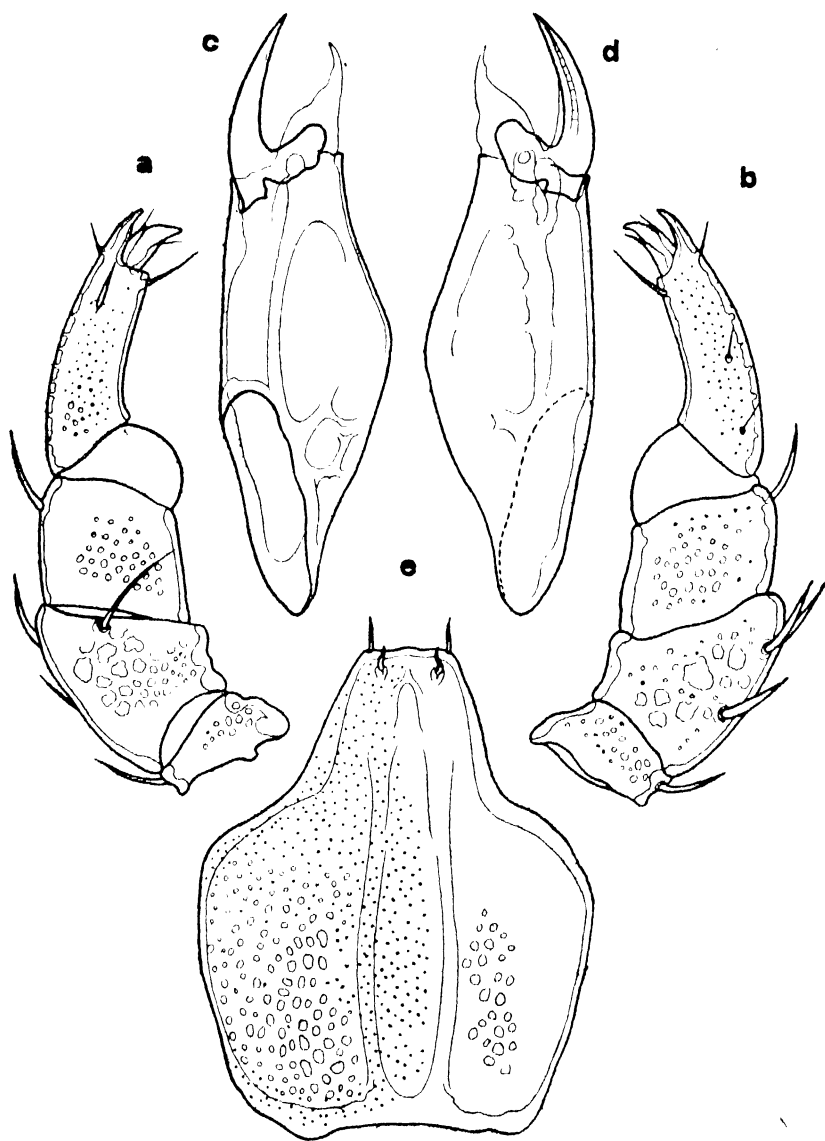
Male. Body large, attaining to the length of 1.33 mm. The general contour tending to develop antero-lateral corners, measuring 1.10 mm wide at the widest anterior region. A slight constriction exists at approximately one third of the total length measured from the anterior extremity. Posterior half encircling a coarsely circular margin. Thickness of the body is considerable, attaining to ca. 1 mm at the genital area. Skin soft, without chitinous plates or platelets. Microscopical appearance of the skin texture is characteristic. The whole surface of the body, except the epimera and the genital area, is covered by numerous small conical papillae whose tips are directed posteriorly. Neighbouring cones show a



Text-fig. 2. *Calonyx japonicus* (UCHIDA). Left, ventral view of male; right, texture of skin from dorsal antero-lateral portion (20×5).

remarkable continuity through radiating ridges which unite with each other (Text-fig. 2, right). Eyes in capsules, one-sided capsule containing double eyes. Maxillary organ broad, being provided with a rostrum which exceeds in length one third of the basal part (Text-fig. 3, e). Mandibles 0.33 mm long, about the same length as palpi and a little longer than the maxillary organ. Claws, measuring 0.08 mm long, stout and gently curved. Hyaline protrusions narrow and considerably elongated. Basal portions broad, widest at a point midway between both extremities, and tapering posteriorly to a pointed end (Text-fig. 3, c & d). Palpi (Text-fig. 3, a & b) thick and porose, the lengths of the palpal segments being (in mm) :

	I	II	III	IV	V
Extensor side	0.02	0.13	0.06	0.16	0.03
Flexor side	0.04	0.04	0.06	0.09	0.02

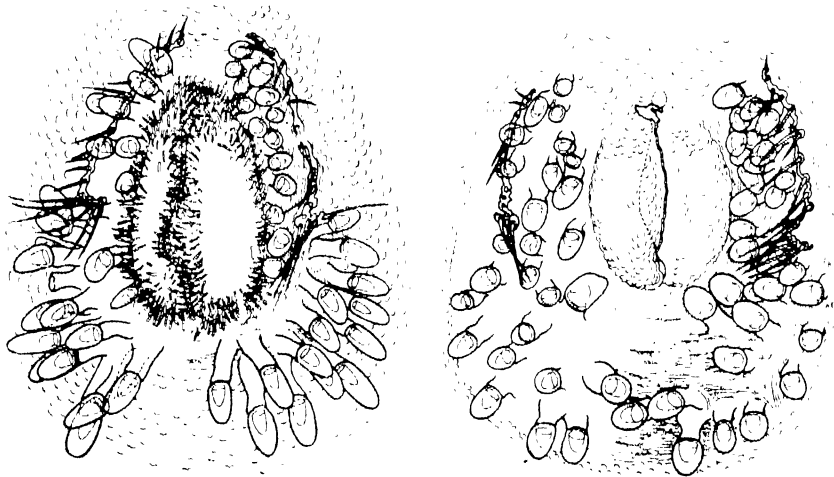


Text-fig. 3. *Calonyx japonicus* (UCHIDA). a, left palpus of male, inner side; b, left palpus of male, outer side; c, dorsal view of mandible; d, ventral view of mandible; e, ventral view of maxillary organ.

Seen from the side the second segment, broadest of all, is of modified trapezoidal form, and is provided with a few bristles. The fourth segment is rather blunt distally, being by no means narrowed in a sig-

nificant manner. Its distal prominence which forms a cheliform structure with the ventrally inserted fifth segment, does not attain to the length of the latter.

Epimera in four groups. They are situated so as to surround the genital area from four opposite directions, the anterior pair taking a close position and the posterior pair being at a distance from each other (Text-fig. 2, left). The first epimera, whose inner edges are uniting with the maxillary base, are of an elongated equilateral triangular form, the tip of which being directed towards the anterior extremity of the genital area. The postero-lateral margins of the second epimera are distinctly concave, thus forming an angle between this concavity and the postero-medial convexity of these plates. The third epimeral plates are separated from the second epimeral plates by a distance which does not exceed the space between the left and right third plates. The inner end of these plates are not pointed nor nearly-pointed, but are continuous in short longitudinal straight lines. The fourth epimera are oblique in position and appear to be of spindle-shape. The inner edge lines of the third and fourth epimera are nearly straight. All the epimeral plates are coarsely porose. Legs porose, devoid of swimming hairs, but a number of moderate bristles in a row is found along the dorsal surface of each leg, especially on the II-V segments. Similar, but more stout, bristles are arranged along the extensor margin of legs, the arrangement being nevertheless sparse. The



Text-fig. 4. *Calonyx japonicus* (UCHIDA). Left, genital area of male; right, genital area of female.

distal segments of all legs end in two strong claws which are characteristic of the other members of the genus *Calonyx*. These claws are of sickle-shape and contain twenty or more teeth, of which the fourth or the fifth counted from the outer side, is most predominant.

Genital area occupying the area of $0.33 \text{ mm} \times 0.26 \text{ mm}$, its outline being pear-shaped, lying almost equi-distant from all the epimera (Text-fig. 2, left). A pair of poorly developed genital plates is present, enclosing the anterior half of the outer margin of the genital area. These plates are represented by only narrow chitinous stripes, which are just wide enough to support the bristles (Text-fig. 4, left). The latter are about 17 in number, inserted along each genital plate, their size increasing in the antero-posterior direction. Inside the genital plates are arranged many acetabula, each stipitate with an elongated oval head. They are planted broadly in two rows on each side, increasing in size posteriorly. The number of acetabula counted in four males is reproduced as follows:

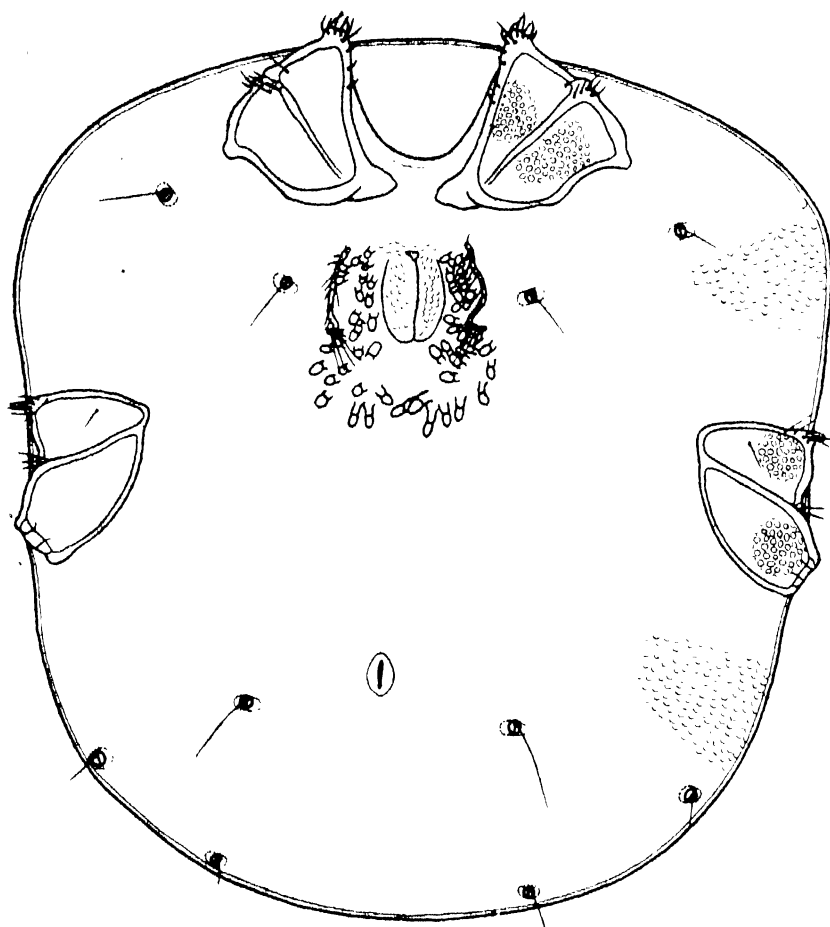
Left	28	22	23	24
Right	25	26	22	27

Immediately around the genital lips, which attain to two-thirds of the length of the genital area, is an eminently convexed genital field. This field is densely covered by numerous short filiform papillae. Outside this field the genital area is smooth-skinned. Excretory pore lying slightly anteriorly to the midway point between the genital area and the posterior body margin.

Colour reddish vermillion. Eyes black.

Female. Far larger than the male, attaining to 2.17 mm long and 1.77 mm wide at the widest anterior portion. The medio-lateral constriction is not remarkable, and the whole body resembles a quadrate whose corners are rounded off. The skin texture, maxillary organ, mandibles and palpi are in good agreement with those of the male. The postero-medial extremities of the first and the second epimera uniting to form a proboscis-like projection; the third and the fourth epimera are like those in the male (Text-fig. 5).

Genital area is situated at the centre of an imaginal trapezoid formed by joining the four groups of epimera. The genital plates are exactly similar to those of the male, bearing about 17 bristles on each plate (Text-fig. 4, right). Number of acetabula shows a considerable variation in different individuals; such as (counted in four females):



Text-fig. 5. *Calonyx japonicus* (UCHIDA). Ventral view of female.

Left	31	21	29	28
Light	26	23	29	30

There is a small chitinous supporting piece at the anterior extremity of the genital lips, around which the skin is furnished with conical papillae like those on other parts of the body, but not with the filiform ones specialized in the male specimens. Colour and eyes same as the male.

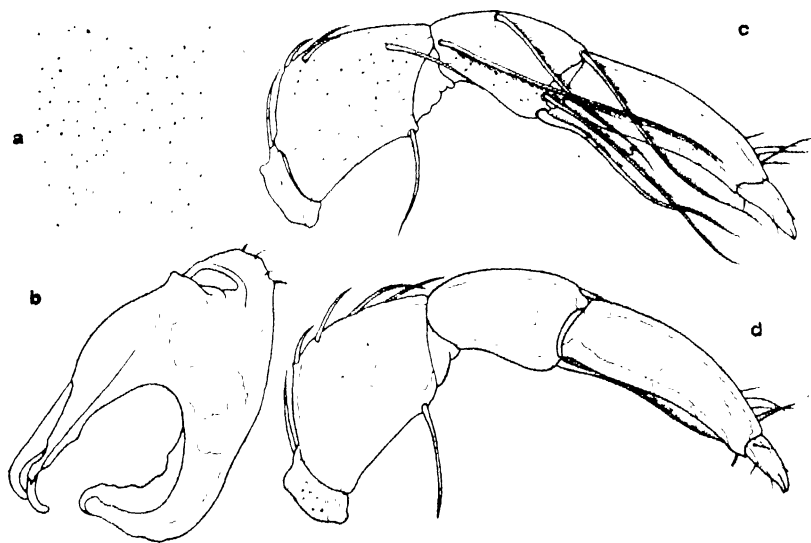
Locality. Four males and four females were collected from beneath stones in a tributary of the River Inôzawa near Yakusu on Jan. 27th, 1939.

Remarks. These specimens are closely related to *Protzia japonica* described by UCHIDA (1934) from Hokkaido, Japan. According to personal communications from Dr. UCHIDA, he is now of opinion that the specimens which were formerly reported as *P. japonica* should be correctly identified as *Calonyx japonicus*, hence this name is adopted also for the present *Calonyx*-specimens. The characteristics described in the present materials show some resemblance to those of *C. rotundus* (WALTER), but are unquestionably distinguished from the latter on account of the presence of a large number of the genital acetabula in both sexes.

2. *Lebertia* (*Pilolebertia*) *leioderma* VIETS

(Text-figs. 6-8)

Male. Body thick, about 0.88 mm of thickness, having smoothly rounded outline, being 1.16 mm long and 1.05 mm wide at the widest middle part. Skin very thin and fragile, furnished with irregularly scattered fine spotlets (Text-fig. 6, a). On the dorsal side no thickened chitinous structure is seen except around the eyes and the respective gland apertures. Gland papillae with accessory hairs arranged in four pairs in two rows on each side. Maxillary organ 0.25 mm long, attaining approximately to two-thirds of length of palpi (Text-fig. 6, b). Mandibles slender and elongated,

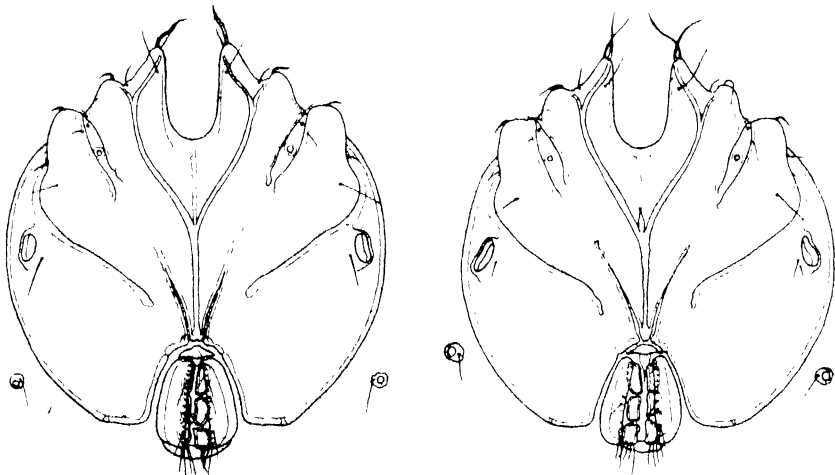


Text-fig. 6. *Lebertia* (*Pilo.*) *leioderma* VIETS. a, texture of skin from dorsal median portion (8×7); b, side view of maxillary organ; c, left palpus; d, right palpus.

slightly exceeding the length of the maxillary organ. Palpi (Text-fig. 6, c & d) of moderate size, elegantly constructed, lengths of segments are (in mm) :

	I	II	III	IV	V
Extensor side	0.03	0.17	0.12	0.16	0.04
Flexor side.....	0.03	0.10	0.08	0.14	0.01

First segment small, with one long bristle at the distal extensor margin. This terminal bristle reaches beyond the proximal as far as one third of the length of the second segment. Second segment, broadest of all, having 0.08 mm of height at the highest portion. Extensor margin well arched, roughly forming two sides of an obtuse angle, being remarkably reflected at about two-fifths from the proximal level. Four bristles implanted along the extensor surface. Flexor side slightly curved ventrally, but is better described as forming a straight line. A long whip-like bristle arises at a point close to the distal end of the flexor margin. Third segment club-shaped, narrowed towards proximal end. Five long feathered bristles arranged on the inner surface, two of them arising from more proximal levels and the other three from the distal margin of the segment. Of these three distal bristles, the one medially located is markedly shifted to the ventral one. All these feathered bristles attain to at least the whole length of the fourth segment, the



Text-fig. 7. *Lebertia* (Pilo.) *leioderma* VIETS. Left, epimera and genital area of male; right, epimera and genital area of female.

most proximal one being the longest. Fourth segment slender, obviously tapering distally, being almost as long as the second segment. Along the flexor edge two hair-pores are present, one being seen at a point midway between the proximal and the distal extremities of the segment, the other at the level of distal quarter. Close to the distal end of the extensor surface are arranged five hairs. Fifth segment ending in two claws. Except the distal and the penultimate segments, all segments exhibiting a tendency to porosity.

Epimera united to form a wide rounded plate covering approximately two-thirds of area of ventor (Text-fig. 7, left). Epimeral plates sculptured with coarse pores, several of them aggregating irregularly. First epimera narrow, extending their tips anteriorly a little beyond the anterior margin of the body. A deep pocket for the maxillary organ is formed between both components of the pair. Posteriorly the first epimera ending in a sharp point which is lying slightly anteriorly to the midway point between the genital area and the maxillary base. Second epimera also narrow, attaining to the genital area by a pair of sharp point ends. Outer sutures between the second and the third epimera vanishing posteriorly far anterior to the inner sutures which are continued just to the anterior extremity of the genital area. Third epimera showing postero-lateral extension at the outer sides, narrowing posteriorly. Suture lines between the third and the fourth epimera do not reach the inner extremity of the plates. Fourth pair, widest of all, with gently rounded sides and slightly concave inner border. Postero-medial ends of the plates are represented by short transverse lines which form an angle of a little over 90° with the longitudinal inner borders. Somewhat condylous thickenings are present at the posterior ends of the second, third and fourth epimera. Legs weak, and rather short as compared with the body. Swimming hairs are found in small numbers (5-10) at distal flexor portion of the Vth segment of the second leg and of the IVth and Vth segments of the third and the fourth legs. Stout spines and bristles arranged around the distal margin and on the dorsal and ventral surfaces of each segment of all legs. Sixth segment ending in two claws, each with two accessory claws and lamina (Text-fig. 8).

Genital area 0.22 mm long and 0.17 mm wide, two-thirds of its length anteriorly being enclosed by the epimeral plates. Sclerites for muscle attachment developed, the posterior one being larger than the anterior one (Text-fig. 7, left). More than twenty hair-pores are seen along the inner edges of genital valves. Genital acetabula are six in number,

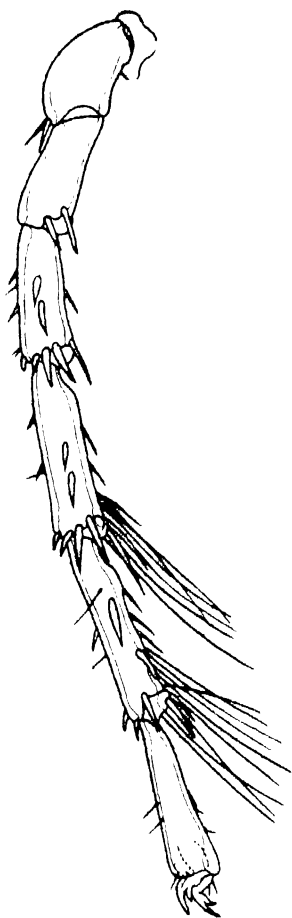
arranged in two longitudinal rows. First pair elongated elliptical; second pair elongated rectangular; third pair smallest with shortened rectangular outline. Excretory pore opens ventrally.

Colour yellowish brown with green patterns. Eyes black.

Female. Body 1.17 mm long and 1.00 mm wide, having thickness of 0.84 mm. Almost all features agree with those of the male, sexual dimorphism being insignificant (Text-fig. 7, right).

Locality. Six males and four females were collected from the main stream of the River Inôzawa, at Otiai on Jun. 8th, 1938.

Remarks. The present specimens show marked resemblance to *L. (Pilo.) leioderma* VIETS reported from Germany. However, closer observation revealed that the distal median feathered bristle of the third palpal segment is more closely shifted to the ventral one than in the German species, and that the hair-pores at the flexor margin of the fourth segment are lying more distally than in the German species. Moreover, the angle of the postero-medial edges of the fourth epimera is greater than 90° in the present materials, and smaller than 90° in the German species. Nevertheless, such morphological disagreements found between the Japanese and the German specimens seem to be insufficient to separate these creatures in different categories.



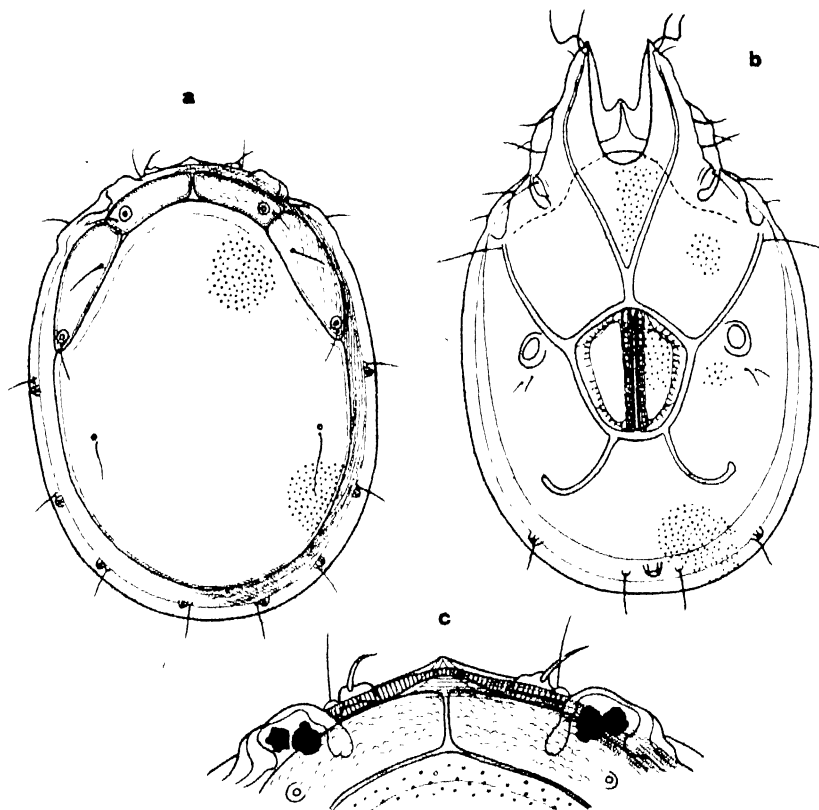
Text-fig. 8. *Lebertia (Pilo.) leioderma* VIETS. Right fourth leg, ventral view.

3. *Atractides (Atractides) ellipticus* (MAGLIO)

(Text-figs. 9 & 10)

Female. Body 0.61 mm in length by 0.44 mm in breadth; dorso-ventral dimension 0.16 mm. Outline elliptical, showing somewhat parallel

lateral borders. Frontal margin between the eyes slightly convex (Text-fig. 9, c). Antenniform bristles poor in development. All of the anterior accessory shields are completely freed from the posterior principal shield which is covering almost the whole surface of dorsum. Anterior two-fifths of the principal shield narrowed to form a blunt protrusion with gently arched anterior extremity (Text-fig. 9, a). Groups of coarse porous



Text-fig. 9. *Atractides (A.) ellipticus* (MAGLIO). a, dorsal view of female; b, ventral view of female; c, anterior frontal portion of female.

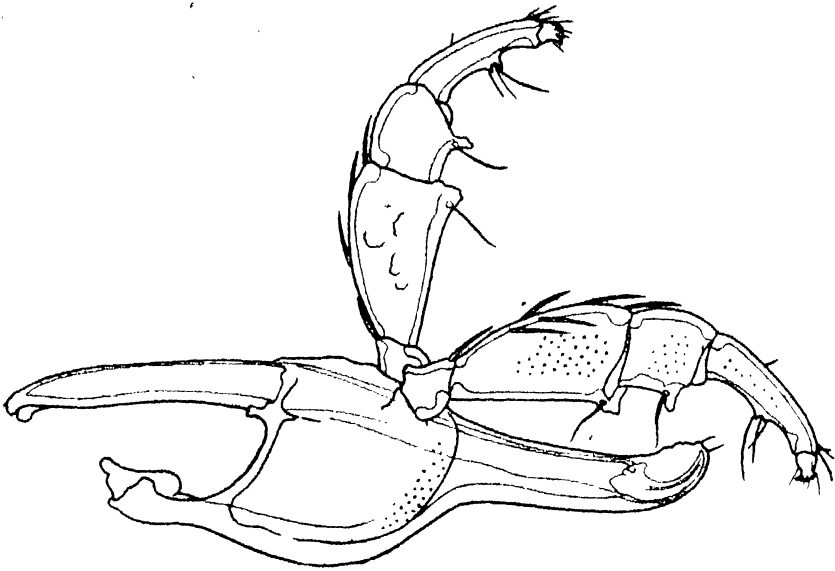
patterns characteristic to the European and the Russian forms are not obviously pointed out on the principal shield. Gland papillae with accessory hairs are very much reduced in number, being represented by a pair of centrally located ones. The anterior pair of accessory shields narrower and smaller than the posterior pair. The latter tapering posterolaterally. Maxillary organ 0.36 mm long, elegantly constructed (Text-fig.

10). Rostrum narrow and elongated, attaining to about 0.15 mm in length; anterior end of rostrum curved dorsally. Basal portion of the maxillary organ extending ventrally, attaining to a height of 0.13 mm at the highest portion. Mandibles elongated, exceeding the whole length of the maxillary organ. Palpi, shorter than the maxillary organ, distinctly narrowed at distal and proximal directions. Outer side of palpal segments, except the distal and the proximal ones, coarsely porose. Measurements given for lengths of each segment are (in mm):

	I	II	III	IV	V
Extensor side	0.04	0.12	0.05	0.10	0.02
Flexor side	0.02	0.10	0.05	0.08	0.02

Second segment, broadest and longest of all, narrowed proximally and forming somewhat wedge-shaped contour. On the gently curved extensor margin are arranged several small bristles. Flexor margin straight, having a small hyaline process and a small bristle at its distal extremity. Third segment short, slightly narrower than the distal half of the second segment. Extensor surface deficient of bristles and hairs. A small hyaline process similar to that of the former segment is seen at the distal extremity of the flexor surface. Fourth segment, narrower than the first segment, rather tapering distally. At approximately the median portion of the flexor edge three minute conical processes are present, each being accompanied by one bristle. Fifth segment, minutest of all, ending in four blunt claws.

Epimera attaining to the length of 0.70 mm, covering almost the whole ventral surface, and extending anteriorly far beyond the anterior body margin (Text-fig. 9, b). Pocket for the maxillary base narrow and deep, developing over anterior half of the first epimera. First epimera ending slightly anteriorly to the genital area, in sharply pointed end. Posteromedial border of the second and the third epimera forming the anterolateral margin of the genital area. Fourth epimera, broadest of all, surrounding the posterior two-thirds of the genital area. Lateral border of the fourth epimera somewhat parallel; posterior border rounded. Legs slender and short, none of them attaining to the length of the body. Swimming hairs absent. Spines and bristles arranged around the distal margin and on the extensor surface of each segment. Last segment ending in two claws, each with two accessory claws and lamina. Genital area 0.18 mm in length by 0.17 mm in breadth, lying at a level two-



Text-fig. 10 *Atractides* (A.) *ellipticus* (MAGLIO). Maxillary organ, mandibles and palpi of female.

thirds the length of the epimera from the anterior end. Genital plates, fringed by numerous minute hairs, developing the antero-lateral corners, and rounded posteriorly. Acetabula twelve in number, arranged in two rows. Excretory pore opens wholly posteriorly near the posterior body margin.

Colour black with yellow streaks. Eyes black, double in capsules.

Locality. One female was secured in the main stream of the River Inôzawa, at Mitukuri, on Jun. 8th, 1938.

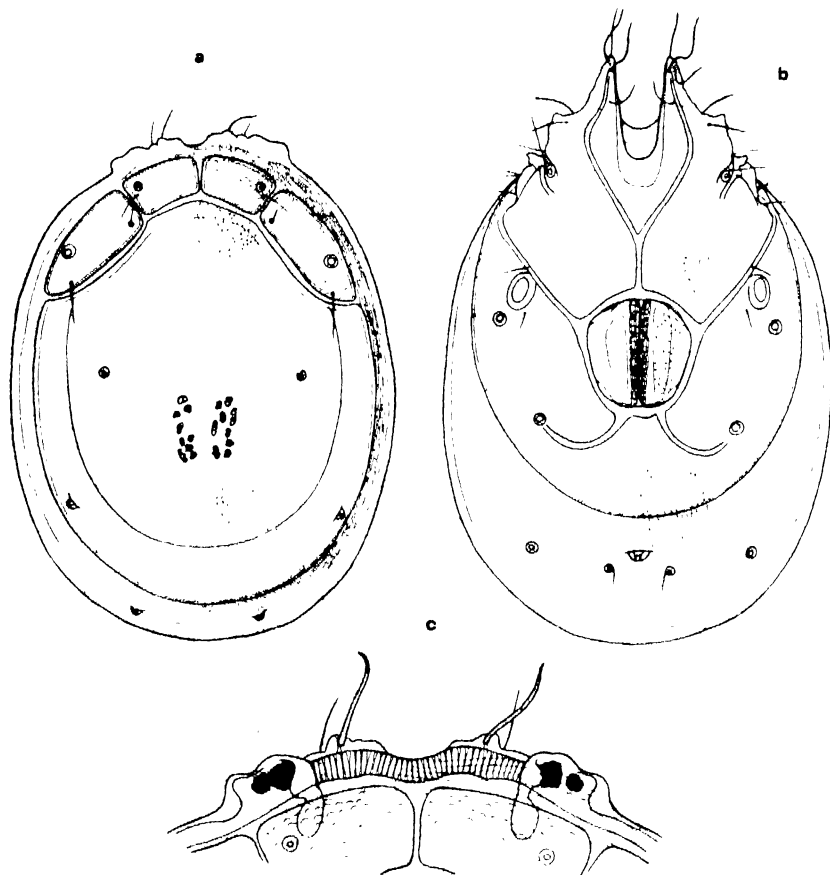
Remarks. The species is an Eurasiatic form widely distributed from Europe to the Ussuri region. The lack of porous pattern for muscle attachment on the dorsal principal shield, and the considerable reduction in the number of gland papillae appear to indicate a slight deviation of the present specimen from the specific characters.

4. *Atractides* (*Atractides*) *stadleri* WALTER

(Text-figs. 11-14)

Male. Body 0.77 mm long excluding the anterior extension of the epimeral plates, and 0.61 mm wide at the widest portion. A slightly rounded oval in outline. Anterior frontal margin between the antenniform bristles distinctly concave (Text-fig. 11, c). Two pairs of anterior accessory

shields are freed from the principal dorsal shield which covers almost the whole dorsal aspect of the body (Text-fig. 11, a). Anterior pair of the accessory shields are smaller than the posterior pair, and having somewhat rectangular outline. Posterior pair connecting to the principal shield along the antero-lateral margin of the latter. At approximately median portion of the principal shield there is a pair of porous patterns said to be the external appearance of structure for muscle attachment. Each



Text-fig. 11. *Atractides (A.) stadleri* WALTER. a, dorsal view of male; b, ventral view of male; c, anterior frontal portion of male.

group of these patterns occupying a narrow elongated area. Maxillary organ broad, with very poorly developed rostrum (Text-fig. 12). Claw of the mandible rather straight. Palpi 0.18 mm long, somewhat stout in appearance. Measurements given for each palpal segment are (in mm):

	I	II	III	IV	V
Extensor side	0.03	0.08	0.05	0.07	0.04
Flexor side.....	0.02	0.05	0.03	0.05	0.03

First segment small, with a short bristle at the distal end of the extensor surface. Second segment, broadest of all, and almost as long as the fourth segment. On the well-curved extensor side are found four short bristles. Flexor surface slightly concave towards proximal end, having an elongated feathered bristle at the distal extremity. Third segment, slightly narrower than the former, having two bristles and a hair on the extensor surface, and a long feathered bristle, as long as that of

the second segment, at the distal end of the flexor surface. Distal margin of this segment distinctly covering the basal portion of the fourth one. Fourth segment short and stout in appearance. A few hairs arising from the extensor surface, and a long bristle and a hair of moderate length are present at the level slightly distal to the middle portion of the flexor surface. The long bristle on the flexor edge more or less exceeding the distal end of the fifth segment. At the distal end of the inner surface of the fourth segment is planted a short strong spine. Distal margin of the same segment has a process on the outer surface and two processes on



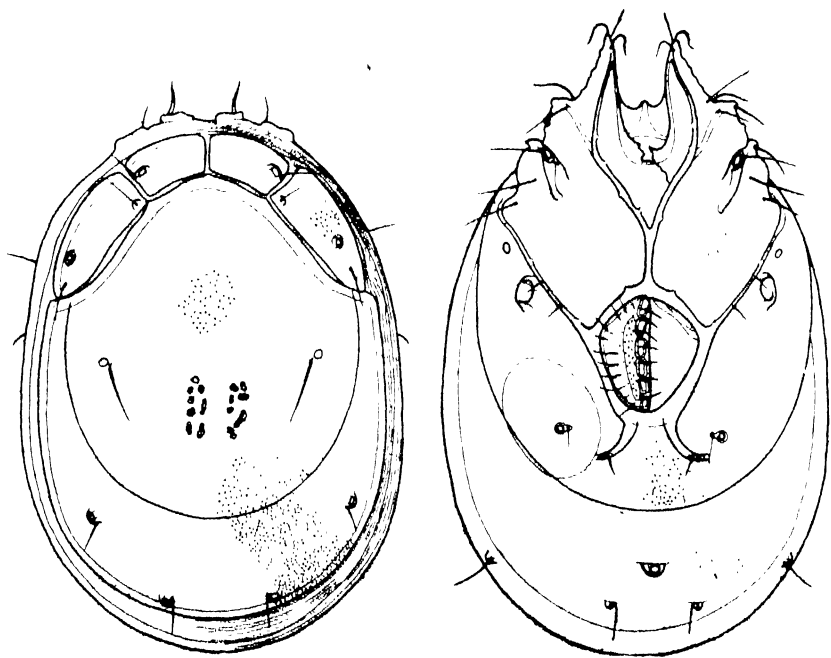
Text-fig. 12. *Atractides (A.) stadleri* WALTER.
Maxillary organ, mandibles and palpi of male.

the inner surface. Fifth segment tapering to four slender claws.

Epimeral region attaining to 0.71 mm long, covering nearly three-fourths of ventor (Text-fig. 11, b). Maxillary pocket narrow and deep, slightly, overlapping one half the length of the first epimera. First epimera end-

ing posteriorly considerably anterior to the genital area. Lateral and posterior border of the fourth epimera markedly rounded. Legs short and slender, none of them being comparable with the body length. Swimming hairs lacking. Several spines and bristles arranged around the distal margin and on the sides of each segment. Distal segment ending in two claws, each with two accessory claws and lamina. Genital area lying broadly at a level two-thirds the length of the epimera from the anterior end. Genital plates wide, shouldered at the antero-lateral edges, and slightly extending laterally at the postero-lateral ones. Acetabula twelve in number, arranged in two rows. Excretory pore opens at a level one third the distance between the posterior border of the fourth epimera and the posterior body margin measured from the anterior end.

Colour black with yellowish white patterns. Eyes black, double in capsules.



Text-fig. 13. *Atractides (A.) stadleri* WALTER. Left, dorsal view of female; right, ventral view of female.

Female. Body oval, widened posteriorly. General contour seeming to be a little narrower than the male. Length, excluding the epimeral protrusion, 0.72 mm by 0.50 mm in breadth at the widest posterior part.

In accordance with the narrowed contour of the dorsal surface, the principal dorsal shield is more or less narrowly elongated in outline as compared with that of the male (Text-fig. 13, left). Maxillary organ 0.18 mm long, broad and with a short rostrum curved slightly downwards (Text-fig. 14). Palpi 0.18 mm long, almost equally long as the maxillary organ. Characteristic arrangement of the feathered bristles at the distal extremity of the second and the third segments, and other features completely agree with those in the male. However, the long bristle on the flexor surface of the fourth segment is rather shorter than that of the male, but overlapping the distal extremity of that segment. Epimera and legs showing close identity to those in the male (Text-fig. 13, right). Genital plates, shouldered at the antero-lateral corners, but narrowed posteriorly, being in the form of triangles. Excretory pore opens at approximately the middle portion of the space between the posterior border of the fourth epimera and the posterior body margin. Colour and eyes same as the male.



Text-fig. 14. *Atractides (A.) stadleri* WALTER. Maxillary organ, mandibles and palpi of female.

Locality. One male and one female were collected at Otiai, from the main stream of the River Inôzawa, on Jun. 8th, 1938.

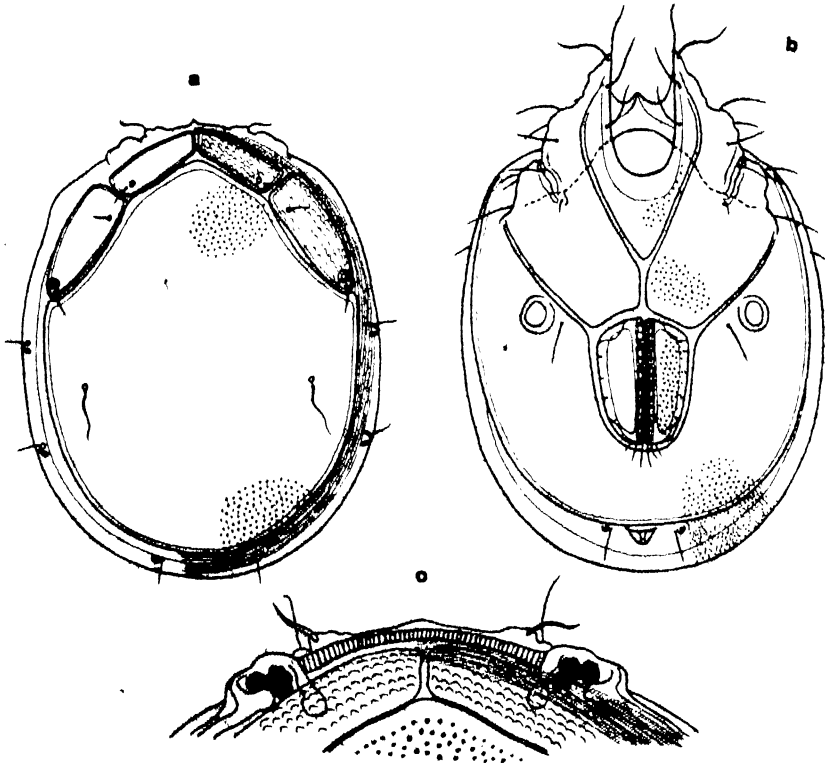
Remarks. *A. (A.) stadleri* WALTER has been reported from Germany and France. The present specimens agree in principal features with the

species above mentioned. However, the presence in the present animals of a long bristle on the flexor surface of the fourth segment seems to indicate a slight variation of them from the European specimens.

5. *Atractides* (*Atractides*) *nipponicus*, n. sp.

(Text-figs. 15-18)

Male. Body elliptical in outline, measuring 0.61 mm long without the anterior extension of epimera, and 0.50 mm wide at the widest portion. Body flattened dorso-ventrally in remarkable way, being 0.16 mm of thickness. Anterior frontal margin between the eyes gently convex (Text-fig. 15, c). Antenniform bristles minute. All the four accessory shields completely separated from the principal one which covers almost the whole surface of dorsum (Text-fig. 15, a). Anterior pair of accessory plates rather smaller than the posterior pair. Posterior pair tapering



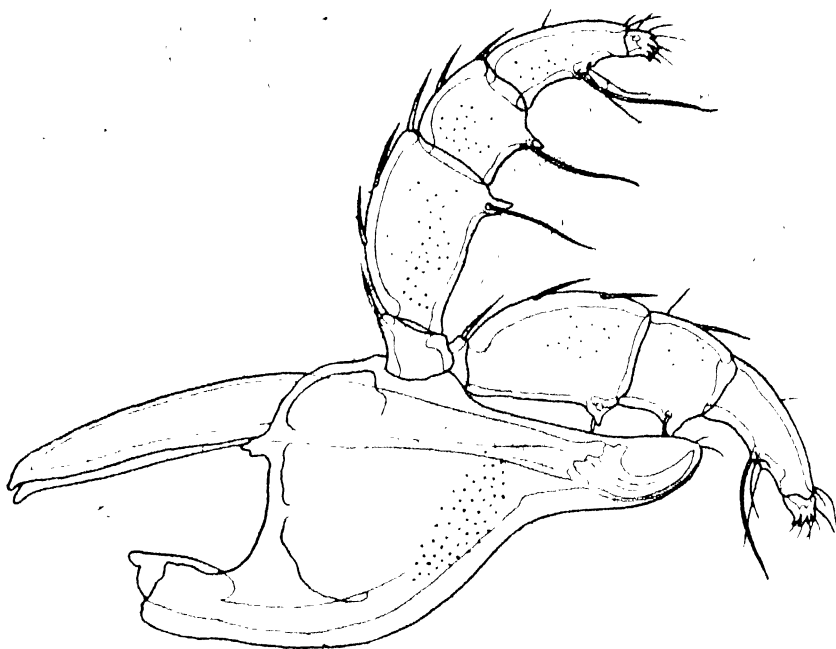
Text-fig. 15. *Atractides* (*A.*) *nipponicus*, n. sp. a, dorsal view of male; b, ventral view of male; c, anterior frontal portion of male.

postero-laterally along the anterior margin of the principal shield. More than one third of the principal shield narrowed anteriorly to form a blunt protrusion, along both sides of which are arranged the accessory shields.

On the principal shield there is no visible pattern of complicated pores, and only one pair of gland papillae with accessory hairs is seen at approximately the median portion of the shield. Maxillary organ appears massive, consisting of a short rostrum and a broad basal portion (Text-fig. 16). Total length of the organ measuring 0.29 mm; rostrum only being about 0.09 mm. Maximal height of the basal portion attaining to 0.16 mm. Mandibles elongated, their posterior parts exceeding the basement of the maxillary organ. Palpi, a little shorter than the maxillary organ, being narrowed at distal and proximal directions. Lengths of each segment are (in mm):

	I	II	III	IV	V
Extensor side	0.04	0.11	0.05	0.09	0.02
Flexor side	0.02	0.09	0.05	0.06	0.03

First segment small, but somewhat high in appearance, having a bristle at the distal extensor margin. Second segment, broadest of all, distinctly narrowed proximally, having gently convexed extensor surface and rather straight flexor edge. A few bristles inserted along the extensor margin, and a small conical hyaline process is found at the distal flexor margin. A long, but slender, bristle arising from the outer base of the process. Third segment short, but almost equally as wide as the former segment, having a few bristles on the curved extensor edge and a small conical process at the distal end of slightly concave flexor margin. A bristle similar to that of the second segment is inserted at the inner base of this process. Fourth segment elongated, but shorter than the second one, being narrowed distally. A minute hair is found on the curved extensor margin, and three bristles, one large and two smaller, arising from a level midway between the two ends of the concave flexor surface, each bristle being inserted at the base of the corresponding small conical process. Fifth segment, minutest of all, ending in four small claws. Epimera covering almost the whole surface of ventor, remarkably protruding anteriorly beyond the anterior body margin. Total length attaining 0.67 mm; maximum width 0.49 mm. Pocket for the maxillary base broad and deep, ending posteriorly at a level three-fifths of the length of

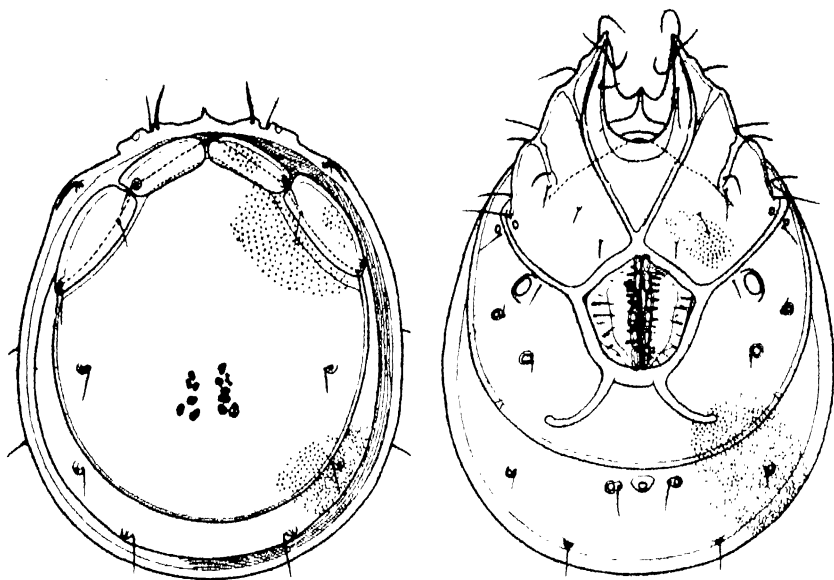


Text-fig. 16. *Atractides* (A.) *nipponicus*, n. sp. Maxillary organ, mandibles and palpi of male.

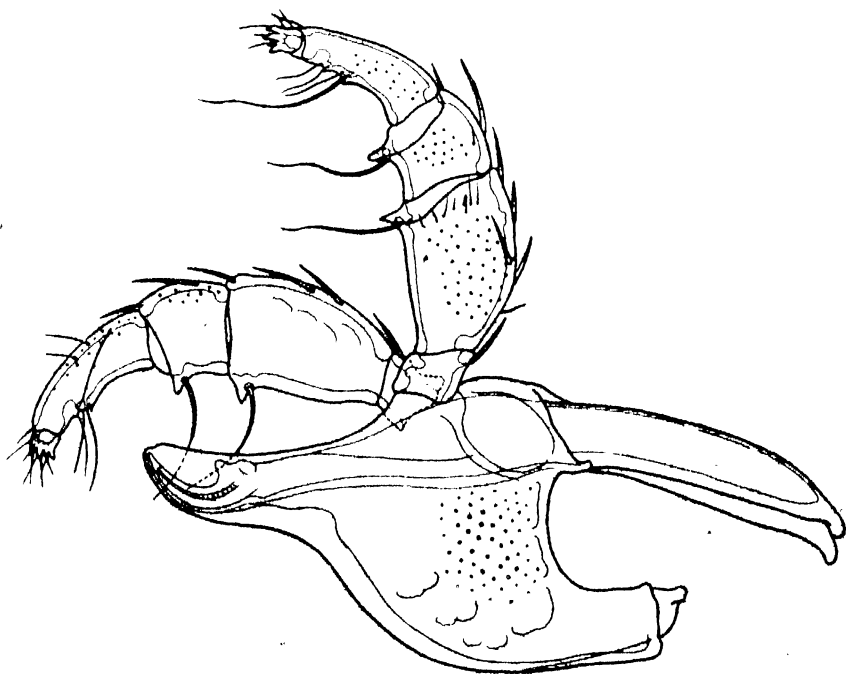
the first epimera measured from the anterior end. First epimera ending posteriorly far anteriorly to the genital area (Text-fig. 15, b). Postero-medial borders of the second and third epimera forming the anterior extremity of the genital area. Fourth epimera, widest of all, completely surrounding the lateral and posterior parts of the genital area. Lateral margins of the fourth epimera gently arched, posterior margins somewhat straight. Legs short as compared with the body. Genital area lying at approximately a level two-thirds the length of the epimera from the anterior extremity, being considerably elongated. Genital plate elongated, slightly widened anteriorly and gradually narrowed to rounded posterior edges. They are fringed by a number of minute protective hairs. Acetabula twelve in two rows. Excretory pore opens far behind the ventral surface, just posterior to the posterior border of the fourth epimera.

Colour black with white patterns. Eyes black, double in capsules.

Female. Outline oval, slightly shouldered at antero-lateral corners. Length 0.66 mm excluding the anterior elongation of epimera; breadth 0.55 mm. On the dorsal principal shield is present a pair of porous patterns at approximately the central portion (Text-fig. 17, left). Number



Text-fig. 17. *Atractides* (*A.*) *nipponicus*, n. sp. Left, dorsal view of female; right, ventral view of female.



Text-fig. 18. *Atractides* (*A.*) *nipponicus*, n. sp. Maxillary organ, mandibles and palpi of female.

of gland papillae greater than in the male. Maxillary organ, mandibles and palpi agree in minutest details with those in the male (Text-fig. 18).

Epimera attaining 0.65 mm long, somewhat shorter in relation to those of the male (Text-fig. 17, right). First epimera elongated to just the anterior extremity of the genital area. Second and third epimera ending postero-medially in oblique-cut end which forms an extended antero-lateral margin of the genital area. Lateral and posterior borders of the fourth epimera rounded. Genital area shorter, but wider than in the male.

Genital plates developing antero-lateral corners and slightly widened at posterior part. Excretory pore opens just posterior to the posterior border of the fourth epimera, and is considerably shifted anteriorly from the posterior body margin. Colour and eyes as in the male.

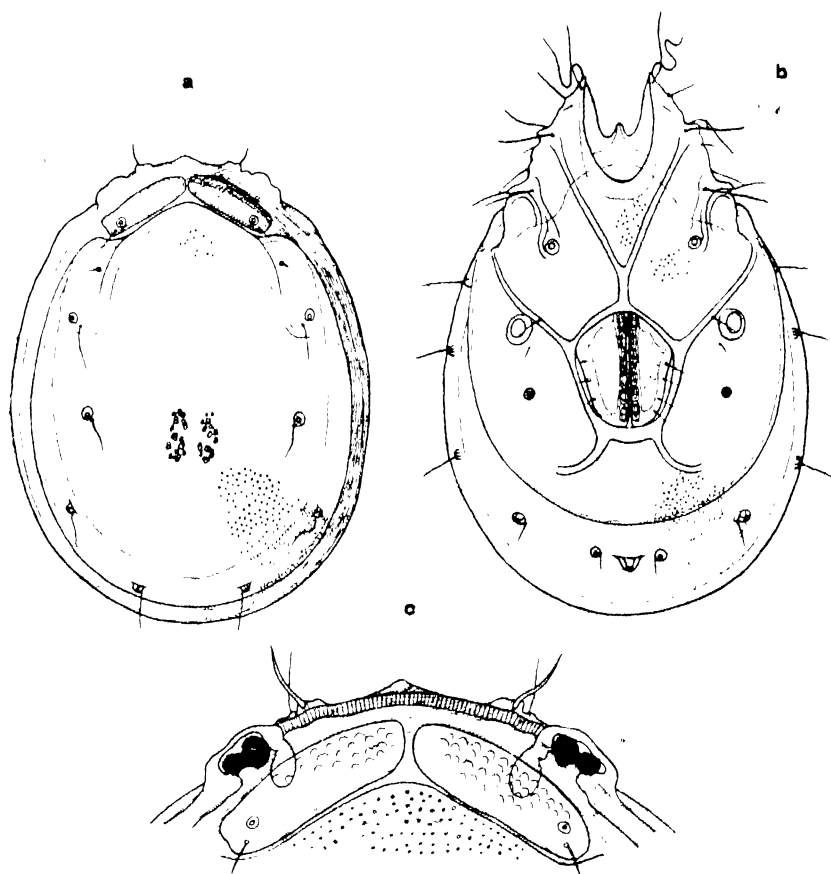
Locality. One male and seven females were collected near Mitukuri, from the main stream of the River Inôzawa, on Jun. 8th, 1938.

Remarks. The present species resembles in several features *A. (A.) brevirostris* HALBERT. However, the facts that the frontal margin of the present specimens has a convex outline, and that the height of the maxillary base exceeds the length of the rostrum, and further that the genital area in the male is distinctly elongated, appear to be enough to separate the present specimens from the species mentioned above.

6. *Atractides (Rusetria) semisutus* SOKOLOW

(Text-figs. 19-22)

Male. Body elliptical, rounded at anterior and posterior extremities, 0.66 mm in length excluding the anterior elongation of the epimera. Maximum breadth 0.55 mm at the median portion of the body. Dorso-ventral dimension attaining approximately one half the maximum breadth. Dorsal surface covered almost entirely by a wide principal shield, having similar contour with the dorsal peripheries, but being a little reduced in its area. Free accessory shields are represented by only a pair of small plates covering the area just anterior to the principal one. Posterior accessory shields fully united to the principal one at antero-lateral corners of the latter. Fused margin between these shields obviously observed for the sake of a subdermal chitinous thickening, which becomes obscure towards posterior direction. On the central median portion of the principal shield is found a pair of porous patterns, each being limited to a rather narrow area and closely approached by the next one (Text-fig.



Text-fig. 19. *Atractides (R.) semisutus* SOKOLOV. a, dorsal view of male; b, ventral view of male; c, anterior frontal portion of male.

19, a). Anterior frontal margin between the eyes gently convex (Text-fig. 19, c). Antenniform bristles poor in development. Maxillary organ 0.36 mm long, longer than palpi, but shorter than mandibles (Text-fig. 20). Rostrum elongated, attaining 0.14 mm in length. Anterior apex of the rostrum slightly elevated dorsally. Mandibles long, 0.43 mm in length, having well-curved claws. Palpi, remarkably narrowed distally, lengths of each segment are (in mm):

	I	II	III	IV	V
Extensor side	0.05	0.12	0.06	0.10	0.02
Flexor side.. ..	0.04	0.09	0.04	0.06	0.02

First segment small, but giving considerable basement to the whole palpus. Second segment, longest and broadest of all, having narrow base, and widened distally. Along the gently arched extensor surface are inserted four bristles and a minute hair. Flexor margin slightly concave at distal half, having a flat triangular process at its distal end. A moderately sized bristle arising from the proximal base of this process. Distal margin of the second segment obviously covering the proximal portion of the third segment. The latter, equally wide as the former one, considerably reduced in its length. Some two bristles are found on the extensor edge, and a small triangular process with a basal moderate bristle at the distal end of the flexor side. Fourth segment narrowed distally, rather insignificant of dimension, having several hairs along the extensor surface. Midway between both extremities of the flexor surface are found a long proximal bristle and two shorter distal hairs, each of them arising from the base of respective small triangular process. Distal



Text-fig. 20. *Atractides (R.) semisutus* SOKOLOV Maxillary organ, mandible and palpi of male.

part of the segment covering dorsally one half the length of the fifth segment. Fifth segment, smallest of all, being represented by four blunt claws with protective hairs. Lateral and posterior borders of the fourth

epimera are circular in outline, extending to the level of the posterior one sixth of the whole length of ventor (Text-fig. 19, b). Receptacular pocket between the first epimera ending posteriorly in distinctly narrowed bay. Anterior tips of the first epimera bearing two pairs of long bristles. First epimera ending posteriorly in a blunt pointed end, considerably anteriorly to the genital area. Second epimera narrowest of all; suture lines between the second and the third epimera attaining to one half the length of the second epimera towards postero-medial direction. Third epimera with warty processes at the antero-lateral corners. Fourth epimera, widest of all, completely surrounding the posterior three-fourths of the genital area. Legs short and slender, anterior three pairs not exceeding the body length. Swimming hairs wholly absent. Several spines and bristles arranged in circlets around the distal margin and the more proximal parts of each segment. Sixth segment of each leg ending in two claws, each of them with two accessory claws and lamina. Genital area lying at approximately central portion of ventor, being of 0.17 mm long and 0.15 mm wide. Acetabula twelve in two rows. Excretory pore opens midway between the posterior border of the fourth epimera and the posterior body margin.

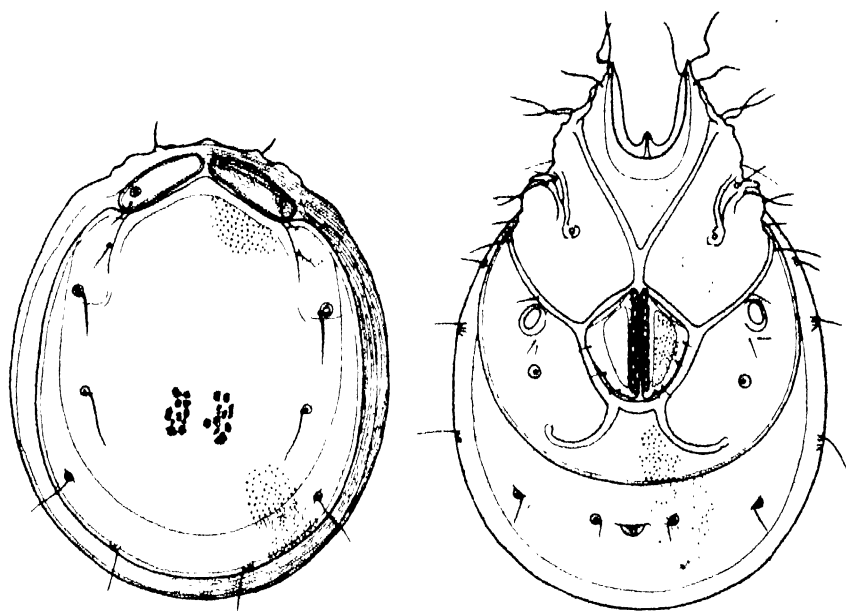
Colour black with yellowish white patterns. Eyes black, double in capsules.

Female. Body the same length as that of the male, 0.66 mm, but slightly wider than the latter, being 0.60 mm at the widest middle portion. Outline oval. In most of the external characters the female specimen agrees with the male (Text-figs. 21 & 22).

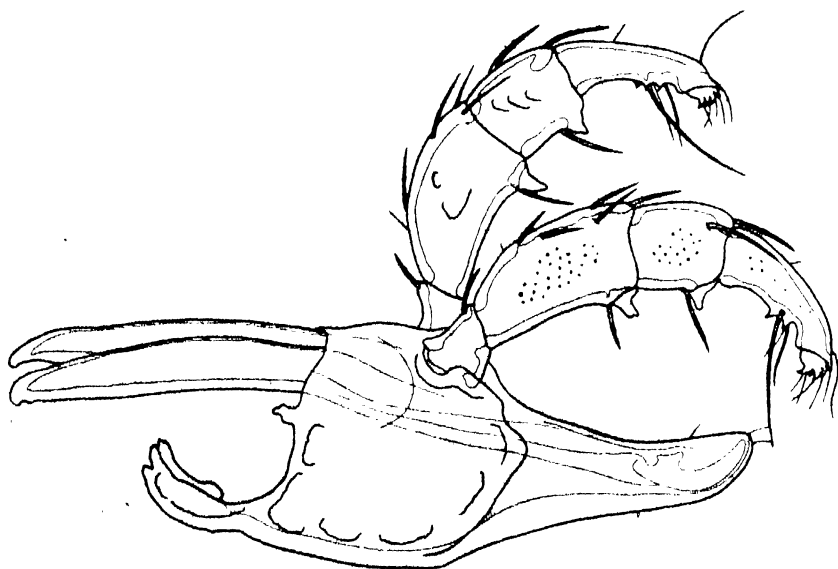
Perceptible differences are pointed out in the form of the receptacular pocket for the maxillary organ, and in the form of the genital plates. The former has a rounded posterior end, wider than that of the male; the latter shouldered at the antero-lateral portions obviously to a greater degree than in the male. Furthermore, the lateral and the posterior borders of the fourth epimera following a pronouncedly rounded outline, being far more rounded than in the male. Colour black with yellowish white patterns. Eyes same as in the male.

Locality. Six males and one female were collected on Jun. 8th, 1938 from the main stream of the River Inôzawa, at O'Kiti-ga-Futi.

Remarks. The species is known in Ussuri region. It shows a close resemblance to *A. (R.) ungeri* SZALAY, but is separable from the latter in having only two bristles at the apex of the first epimera. The present Japanese specimens agree with SOKOLOV's description, except for a long



Text-fig. 21. *Atractides (R.) semisutus* SOKOLOV. Left, dorsal view of female; right, ventral view of female.



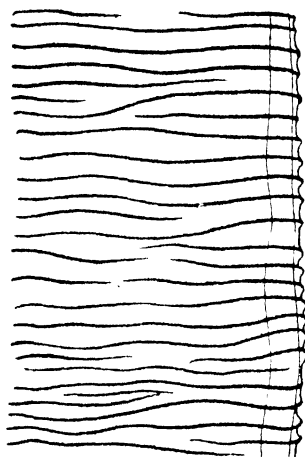
Text-fig. 22. *Atractides (R.) semisutus* SOKOLOV. Maxillary organ, mandibles and palpi of female.

bristle at the middle portion of the flexor surface of the fourth palpal segment in the Japanese specimens.

7. *Hygrobates (Hygrobates) calliger* PIERSIG

(Text-figs. 23-25)

Male. Body rounded in outline, slightly extending anteriorly. Length



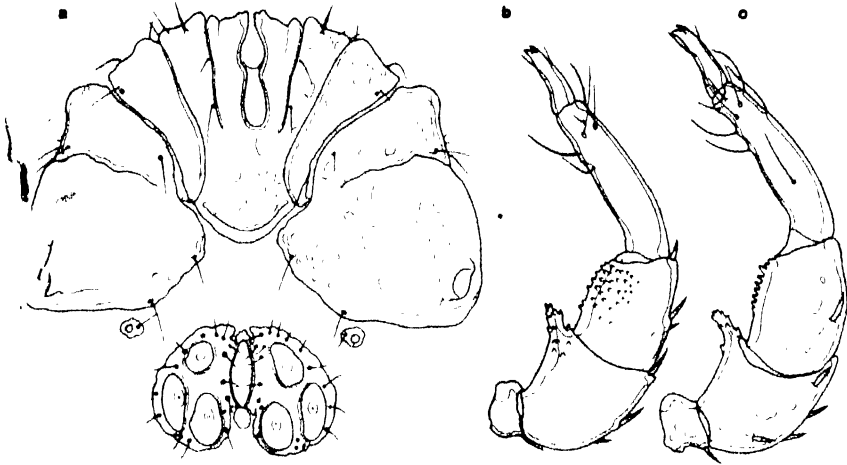
Text-fig. 23. *Hygrobates (H.) calliger* PIERSIG. Texture of skin from dorsal antero-lateral portion (20 \times 5).

0.66 mm; breadth 0.55 mm at the widest middle portion. Body dorsally well-arched, dorso-ventral dimension being 0.44 mm. Venter almost flat. Skin decidedly thin, fragile, apparently finely ridged with transverse parallel ridges which are observed as narrow, pointed elevations along the peripheral margins (Text-fig. 23). On the dorsal side no chitinous thickening is present except around the gland apertures. Maxillary organ fused with the first pair of epimera in the posterior half, measuring 0.10 mm in breadth at the anterior extremity. Posteriorly the basal portion of the maxillary organ somewhat narrowed (Text-fig. 24, a). Mandibles short. Palpi (Text-fig. 24, b & c) more than one third of body length, segments measuring (in mm):

	I	II	III	IV	V
Extensor side	0.03	0.12	0.09	0.13	0.05
Flexor side.....	0.04	0.06	0.06	0.10	0.05

First segment short, having a minute bristle at the distal end of the extensor surface. Second segment, broadest of all, and widened distally, with rounded extensor margin and concave flexor one. Along the extensor surface several small bristles arranged in alternate position. From the distal extremity of the flexor surface a high process arises with narrow base, being covered with coarse chitinous denticles which are enriched on the outer side. Third segment of almost equal breadth as the former one at its proximal end, slightly narrowed distally. Flexor surface somewhat convex, with distal two-thirds of area covered by a number of

chitinous denticles which are chiefly delimited on the outer surface of the segment. Extensor surface provided with a few bristles similar to those of the second segment. Fourth segment, longest of all, narrowing



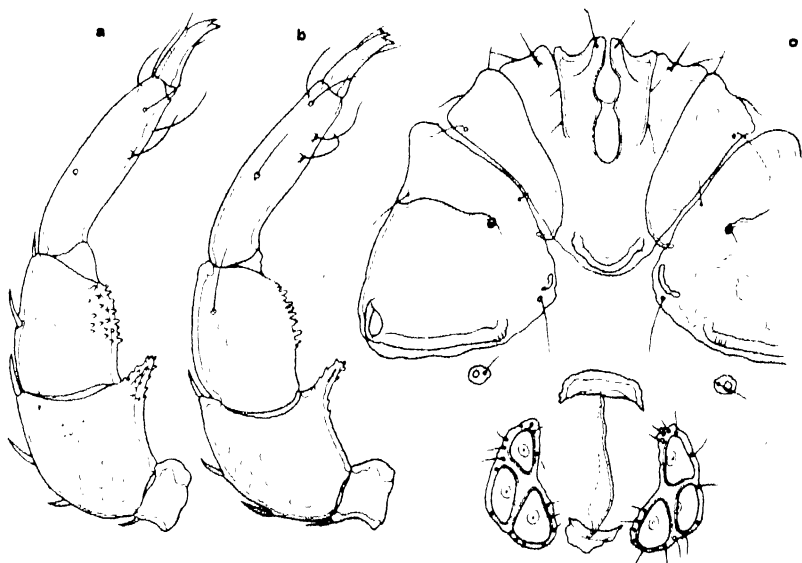
Text-fig. 24. *Hygrobatos (H.) calliger* PIERSIG. a, epimera and genital area of female; b, left palpus of male; c, right palpus of male.

ceptibly at both ends, having two ventral hairs on the distal half of flexor margin, several hairs around the distal portion, and a long at the proximal portion, at one third of the level on the inner surface. Fifth segment ending in two claws and two hairs. Epimeral region micircular in rough contour, covering more than two-fifths of length of center (Text-fig. 24, a). First pair united with the maxillary organ on the inner posterior half, having outer edge confused with the second pair in the anterior half, and expanded out in the posterior half over the second. Posterior extremity of the first epimera forming gently rounded protrusion. Second pair, shorter than the first, united obliquely to the first one, separated from the third one by a narrow interval. Third pair, shorter than the second one, nearly wedge-shaped, tapering towards inner end. Suture lines between the third and the fourth epimera do not reach the inner extremity of the plates formed by fusion of these two pairs. Fourth pair, largest of all, being almost pentagonal in form. lateral margins tending to develop postero-laterally. Inner end of the plate modified into a blunt process. Between these processes on both sides occupying a moderately widened space. Posterior borders of the plates show transverse straight lines. Along the anterior and posterior

corners of the inner margins are present thickened subdermal each bearing a hair. Legs slender, wholly destitute of swimming. Around the distal margin and on the dorsal and ventral surf arranged longer or shorter bristles. Genital area lying nearly central portion of ventor, occupying the length of 0.13 mm and the width of 0.18 mm. Anterior portion of genital area is found in a position slightly posterior to the posterior border of the fourth epimera. At the anterior and posterior ends of the genital aperture are respectively two small chitinous sclerites for muscle attachment. Genital plates uniting the extremities, surrounding the genital lips. Posterior to the genital aperture is present a deep bay freed from the genital plate. The shape of a genital plate resembles a cocoanut. Acetabula three in number on each genital plate, arranging roughly at three angles of a triangle, anteriorly, the other two being shifted posteriorly. Genital plates are fringed by a number of minute hairs. Excretory pore opens midway between the genital area and the posterior body margin.

Colour brown. Eyes black.

Female. Female almost equal in size to the male, attaining to 0.6 mm in length and 0.61 mm in width. It differs from the male only in the form of the genital area (Text-fig. 25, c). In this sex, the genital aperture



Text-fig. 25. *Hygrobatos (H.) calliger* PIERSIG. a, right palp of female; b, left palp of female; c, epimera and genital area of female.

is demarcated at both extremities by two crescent-shaped sclerites for muscle attachment, each of them attaining to a considerable size, the anterior one being larger than the posterior one. Genital plates widely separated from the aperture, each having elongated outline resembling the shape of a bean. Acetabula arranged in alternate positions roughly along the longitudinal direction. Eggs spherical, 0.11 mm in diameter. Colour and eyes same as in the male.

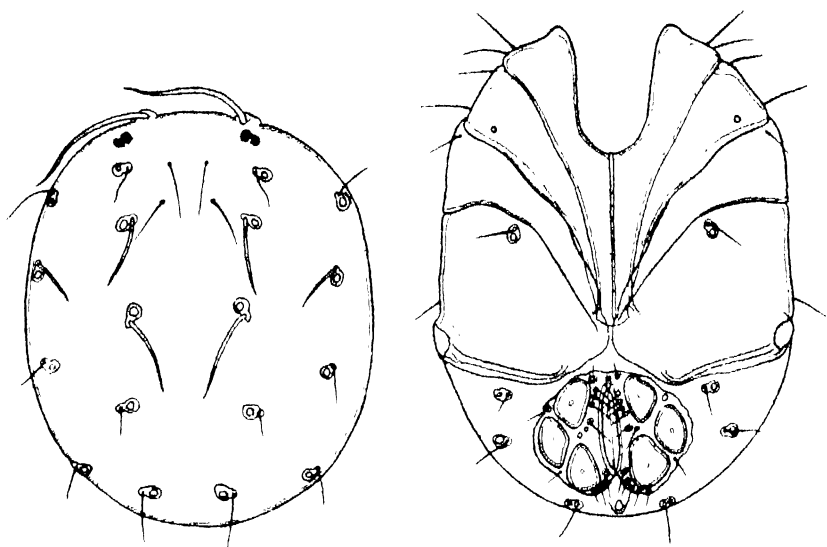
Locality. Eight males and nine females were secured along the course of the main stream of the River Inôzawa on Jun. 8th, 1938.

Remarks. The species is known in various places in Europe. Although the present specimens generally agree with the European materials, they appear to differ from the latter in a slight degree in the form of the fourth epimera and of the male genital area; that is, the present specimens show the postero-lateral portion of the fourth epimera slightly extended outwards, and the posterior furrow of the male genital area deeper than that of the European forms.

8. **Megapus** (**Megapus**) **izuensis**, n. sp.

Text-figs. 26-29:

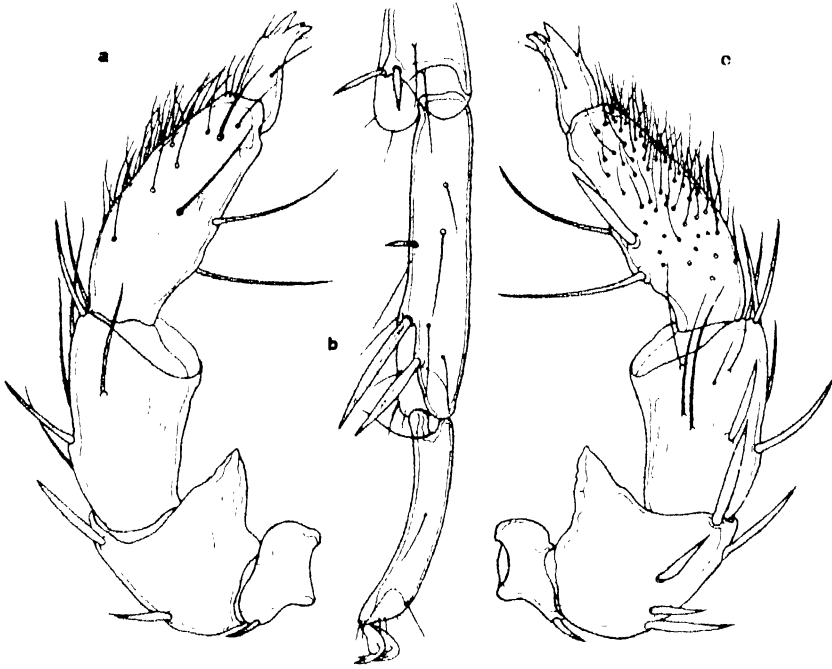
Male. Body small, being 0.33 mm long and 0.27 mm wide at the widest portion. Outline short elliptical, obscurely developing antero-lateral and postero-lateral corners (Text-fig. 26, left). Anterior edge-line somewhat transversely straight; posterior margin well arched. On the dorsal surface are not observable any chitinous plates nor platelets, except an insignificant thickening of chitin around the gland papillae. Eyes double, do not appear to be enclosed in capsules. Skin thin and fragile, without any distinct structures. Antenniform bristles stout and considerably elongated. Gland papillae with respective accessory hairs and bristles arranged in four rows, the outer ones consisting of four papillae and the inner ones of five. Bristles accompanying the second and the third papillae of the inner pair of these rows are very significant. Maxillary organ 0.13 mm long; rostral portion is much reduced. Mandibles 0.22 mm long, their basal portion being prolonged posteriorly far beyond the posterior extremity of the maxillary base. Claws large, falciform, being well curved dorsally. Palpi (Text-fig. 27, a & c) almost equally as long as the mandibles. Measurements for the palpal segments are (in mm):



Text-fig. 26. *Megapus* (*M.*) *izuensis*, n. sp. Left, dorsal view of male; right, ventral view of male.

	I	II	III	IV	V
Extensor side	0.03	0.09	0.08	0.10	0.04
Flexor side	0.03	0.04	0.05	0.08	0.04

First segment small, with distinctly concave extensor surface and well-arched flexor surface. A small bristle is inserted at the distal extensor margin. Second segment, broadest of all, developing its extensor side remarkably. A conspicuous curvature is found between the proximal rounded margin and the distal gentle curvature at approximately the level of proximal one third of length along the extensor edge. Five spines arranged in alternate positions on both sides of the extensor margin. Flexor surface much reduced in its length in relation to the extensor one, having distally a massive triangular chitinous process whose basal portion occupies about two-thirds of length of this side. Height of this process is considerable, attaining approximately 0.02 mm, corresponding to more than one third of height of the segment proper at the highest portion. Third segment narrower than the former one at the proximal end, but widened at its distal end to attain to a height almost comparable with that of the latter. Extensor surface perceptibly convex; flexor surface obviously concave. Outer surface of the segment is supplied with only



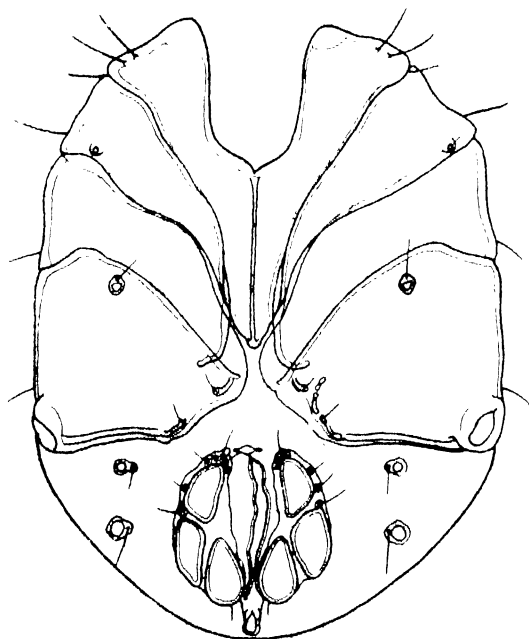
Text fig. 27. *Megapus (M.) izuensis*, n. sp. a, right palpus (outer side) of male; b, IV-VI segments of left first leg of male, ventral view; c, right palpus (inner side) of male.

one bristle arising from the point near the distal margin. Along the extensor side several stout or slender bristles are arranged, three of which aggregating at the distal margin. Several long slender bristles are inserted on the distal half of the inner surface. Fourth segment, longest of all, gradually narrowed towards proximal and distal extremities. Extensor side gently curved; flexor surface has concavity near proximal and distal ends, concavity at the proximal position being deeper than that at the distal one. On the inner surface of the segment, especially on the dorsal half of this surface, are present a number of minute hairs which are very sparse on the outer surface. A strong, but not too long, spine is arising near the flexor edge at approximately the median portion of the inner surface, and two long bristles are shown on both sides of, and close to, the flexor edge-line, the one on the inner surface being at the level of proximal one third of full length of the flexor margin, another one on the outer surface at the level of proximal three-fifths. At about the middle portion of the outer surface is present a long bristle, and

further distally is another one. Fifth segment tapering distally, ending in three claws, one dorsal, the other ventral in position.

Epimeral region very wide, covering almost three-fourths of the ventral area (Text-fig. 26, right). First pair narrow in longitudinal direction, forming a deep pocket for the maxillary organ between the anterior half of both-sided components. Posterior extremity of the first epimera ending at the level of anterior three-fifths of venter, a little anteriorly to the genital area. Second pair attached obliquely to the former, tapering posteriorly to a terminal portion common to the second pair and the first pair. Fourth epimera, broadest of all, having somewhat pentagonal outline, being cornered at antero-lateral, antero-medial, postero-lateral and postero-medial portions. Along the posterior margin is present a subdermal thickening. Medial extremity together with the posterior end of the third epimera forming a blunt protrusion just posterior to the common termination of the first and the second epimera. Both-sided processes show very close approach, leaving almost no space between the two. Legs long in relation to the body dimensions, the posterior legs being the

longest. First pair slightly longer than the body length; fourth pair nearly twice in length of the latter. Swimming hairs absent in all legs. Spines and bristles are densely arranged around the distal margin and at various intermediate levels of each segment in roughly circular arrangement. Fifth segment of the first pair elongated, having double border at distal one third of the flexor side (Text-fig. 27, b). Two long spear-shaped spines are inserted on the ventral flexor margin, proximal one obviously longer than the distal



Text-fig. 28. *Megapus* (*M.*) *izuensis*, n. sp. Ventral view of female.

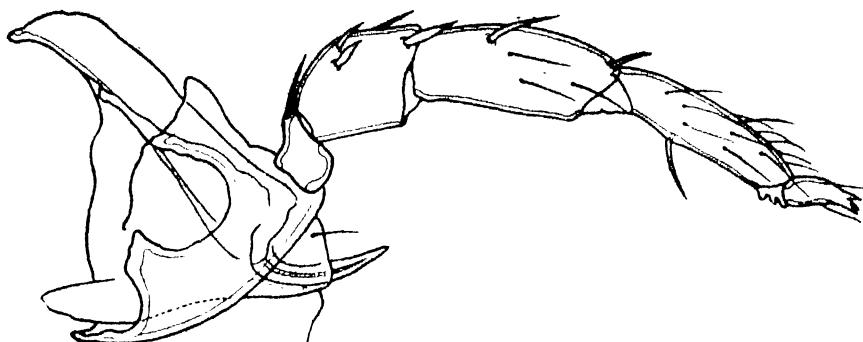
one. Sixth segment of the first pair curved, but not too strongly. Terminal segment of each pair ending in two small claws, each being furnished with two accessory claws and poorly developed lamina. Genital area significantly extended as compared with the whole ventral surface, occupying 0.11 mm long and 0.14 mm wide. Anterior portion of the genital area a little extending into a shallow bay formed by the postero-medial borders of the fourth epimera (Text-fig. 26, right). Posterior extremity of the genital area close to the posterior margin of the body. Genital aperture lying longitudinally between the genital plates, the latter being fused at anterior and posterior ends of the aperture. Acetabula three on each genital plate, being arranged alternately on somewhat triangular base. A number of minute hairs are present around the acetabula, especially along the inner margin of the plate. Excretory pore opens ventrally just posterior to the genital area.

Colour brownish black. Eyes black.

Female. Body measuring 0.50 mm in length by 0.39 mm in breadth; dorso-ventral thickness 0.33 mm. Outline elliptical and slightly cornered at antero-lateral and postero-lateral portions, being similar to the male. Dorsal features, maxillary organ and mandibles show allied configurations to those in the male. Palpi (Text-fig. 29) with uniformly elongated slender segments. Lengths of each segment are (in mm):

	I	II	III	IV	V
Extensor side	0.04	0.10	0.13	0.12	0.04
Flexor side	0.05	0.05	0.10	0.10	0.05

First segment small, having a bristle at the distal extremity of the extensor margin. Second segment rather short, showing well-arched extensor margin and straight flexor margin. Some four bristles are present on both sides of the extensor edge. Third segment, longest of all, having almost uniform thickness along the whole length. Extensor margin gently curved, and flexor margin nearly straight. A few bristles and hairs arranged on the distal half of the segment. Fourth segment equally as long as the former, slightly narrowing distally and proximally. A number of hairs planted on both sides of the extensor margin, and a short bristle is found on the flexor margin at the level of proximal quarter of the whole length of the segment. Fifth segment small, tapering in three claws. Epimera covering approximately two-thirds of venter; their arrangement and contours similar to those in the male (Text-fig. 28).



Text-fig. 29. *Megapus* (*M.*) *izuensis*, n. sp. Maxillary organ, mandible and right palpus of female.

Genital area lying a little apart from the posterior margin of the fourth epimera. Anterior to the genital aperture is developed a small chitinous sclerite for muscle attachment. Acetabula three on each genital plate which are separated from each other. Excretory pore opens just posterior to the genital area. Colour and eyes as in the male.

Locality. One male and one female were secured from the main stream of the River Inôzawa, at Mitukuri, on June 8th, 1938.

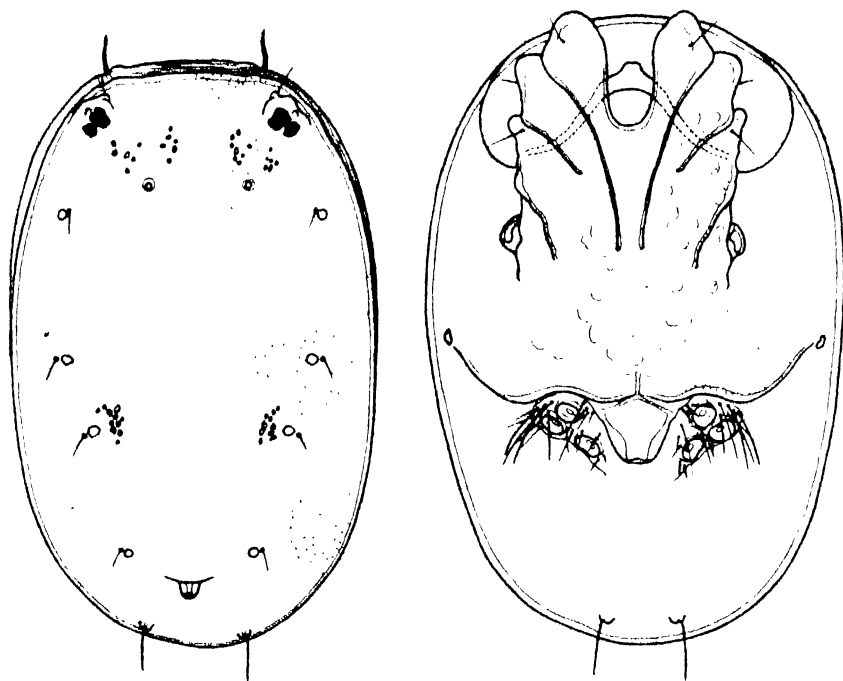
Remarks. The present specimens show a slight resemblance to *M.* (*M.*) *nodipalpis* THOR in the form of palpi, but are entirely different from the latter in many characteristic features in the specialized palpal segments, epimeral forms and also in the genital region. The materials appear to be new to science, hence a new category may be offered for them.

9. *Brachypoda versicolor* (MÜLLER) var.

(Text-figs. 30-33)

Male. Body elliptical in outline, terminating anteriorly in a somewhat truncated edge. Lateral margins parallel, posterior portion smoothly rounded. Length 0.50 mm, breadth 0.33 mm at the widest middle portion. Body fully compressed dorso-ventrally, its thickness being 0.11 mm. Almost the whole surface of dorsum is covered by a dorsal shield which has an outline completely fitted to the general contour of the body (Text-fig. 30, left). Eyes double, shifted far anteriorly close to the antero-lateral corners of the dorsal shield. On the dorsal shield are arranged five pairs of gland apertures, each with respective accessory hair. Immediately before

the eyes are present a pair of minute hairs on small conical ridges. Surface of the dorsal plate is sculptured by minute pores, several of

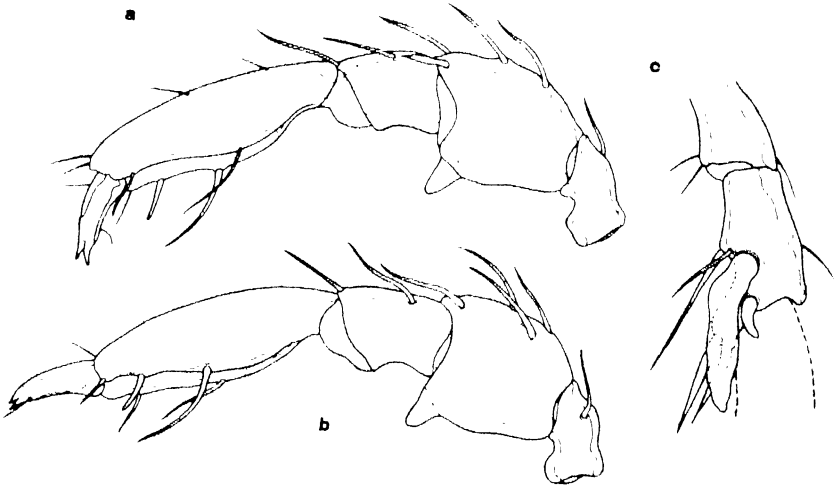


Text-fig. 30. *Brachypoda versicolor* (MÜLLER) var. Left, dorsal view of male; right, ventral view of male.

them aggregating to form groups. At anterior and antero-lateral peripheries the plate is evidently separated from the ventral plate by a narrow non-chitinous groove. The groove is finely striated with parallel ridges of skin. Antenniform bristles not very large, being inserted at the antero-lateral edges. Porose sculpture of the dorsal shield is specially differentiated into three pairs of groups of large pores, of which two are found at approximately the same level behind the eyes and the rest at the level of posterior three-fifths of the shield. Maxillary organ 0.09 mm long, does not reach one half of length of palpi. Mandibles small, being of the same length as the maxillary organ. Palpi (Text-fig. 31, a & b) long, nearly half the length of the body. Lengths of each palpal segment are (in mm):

	I	II	III	IV	V
Extensor side	0.04	0.07	0.04	0.10	0.03
Flexor side	0.03	0.05	0.02	0.08	0.03

First segment narrow but moderately elongated, with one bristle on the extensor surface. Second segment, broadest of all, showing gently arched extensor margin and perceptibly convexed flexor margin. About five bristles are arranged along the extensor surface, and a prominent hyaline process is attached at the distal extremity of the flexor edge. Third segment almost equal in length to the first one, but thicker than the latter, having slightly curved extensor edge and well-arched flexor margin. Two bristles, one proximal and one distal, are inserted on the



Text-fig. 31. *Brachypoda versicolor* (MÜLLER var. a, right palpus of male; b, left palpus of male; c, III & IV segments of left fourth leg of male, ventral view.

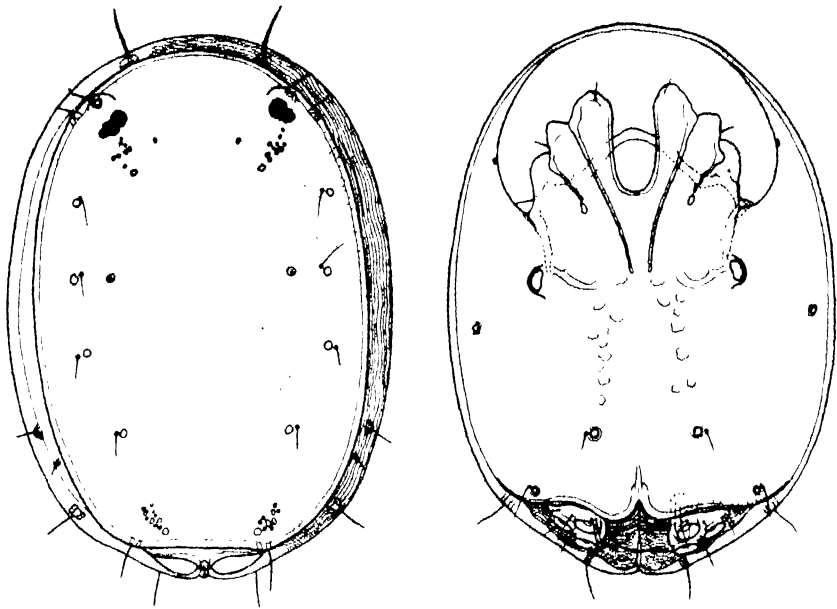
extensor surface. Fourth segment, longest of all, remarkably narrowed proximally. On the flexor side the margin is doubled, each edge-line having two long bristles. Of these bristles those present on the outer edge-line are stronger than those on the inner one. A few minute hairs are seen along the extensor margin and at the distal end of the segment. Fifth segment tapering to two claws.

Epimera unite to form a wide plate which covers about three-fifths of venter (Text-fig. 30, right). First epimera a little extending anteriorly beyond the anterior margin of the body, enclosing a deep pocket for the

maxillary organ. Suture lines between the first and the second epimera are prolonged posteriorly to about half the length of the epimeral plate. Second epimera small, attaching obliquely to the postero-lateral margin of the first ones. Suture lines between the second and the third epimera short. Third epimera attaching just posteriorly to the former ones and markedly oblique in position. Fourth epimera, widest of all, their posterior border constituting a transverse wavy line, extending antero-laterally with arched curvature. At the termination of this border line there is a pair of large pores. Legs (right fourth leg and the Vth and VIth segments of left fourth leg are injured) short in relation to dimensions of the body, the anterior three pairs not reaching the full length of the body. Scanty arrangement of the swimming hairs is observed at the distal margin of the IVth and Vth segments of the second pair of legs, and at the distal margin and on the flexor surface of the IIIrd to Vth segments of the third pair. Spines and bristles arranged around the distal margin and on the extensor surface of each segment. Fourth segment of the fourth pair is sexually characterized (Text-fig. 31, c). This segment is distinctly narrowed at the flexor side at its distal half, the distal part being prolonged distally towards the flexor side beyond the distal extremity of the extensor side. This distal prominence ends in two strong sharp spines. Out of these spines arises a large horn-shaped spine from the flexor surface at a point midway between the distal and the proximal extremities of the segment. Further, a small claw-shaped spine is inserted on the ventral surface, slightly distally to the large one. A few bristles are seen around the middle portion of the segment. Genital area situated close to the posterior border of the epimeral plate, genital aperture opening at the posterior end of a truncated conical process which has its base directly on the epimeral border. On both sides of this genital process are arranged three acetabula in triangular position on a genital plate. Of these acetabula the anterior and the antero-lateral are closely located, and are a little apart from the postero-medial one. Surrounding these acetabula are set many hairs. Excretory pore opens dorsally, near the posterior body margin.

Colour purple. Eyes black.

Female. Body elliptical, anterior portion rounded, posterior margin slightly narrowed. Length 0.55 mm, breadth 0.38 mm at the widest middle part. Body decidedly thick, attaining to about 0.22 mm in thickness. Dorsal shield porose like that of the male, having elliptical outline except the posterior margin which is replaced by a transverse edge



Text-fig. 32. *Brachypoda versicolor* (MÜLLER) var. Left, dorsal view of female; right, ventral view of female.

(Text-fig. 32, left). The characteristic groups of large porous patterns on the dorsal shield of the male are represented in female by two pairs of groups distributed immediately posteriorly to the eyes and near the posterior extremity of the shield. Around the dorsal shield is a wide band of the dorsal groove, on which are situated a few pairs of gland papillae with accessory hairs. Skin of the dorsal groove is finely striated with undulating ridges. Maxillary organ, mandibles and palpi (Text-fig. 33), all agree in minutest details with those of the male. Epimera united to form a broad ventral plate covering almost the whole area of ventor save the posterior triangular portion which is non-chitinous and occupies but one eighth of the length of ventral aspect (Text-fig. 32, right). The three anterior pairs of epimera are far removed posteriorly from the anterior margin of the body, the anterior prominence of the first pair arising at a point one eighth of the level of ventor measured from the anterior. Features of epimera roughly agree with those in the male. Posterior border of the fourth epimera transversely waved, extending perceptibly antero-laterally at both sides. Legs short, none of them exceeding the body length. Swimming hairs are present sparsely at the distal margin of the Vth segment of all the pairs. Bristles and

spines arranged in circlets around the distal end, and on the extensor and the flexor margins of each segment. Fourth segment of the fourth pair elongated, being furnished with several simple bristles around its margin. Fifth segment slender, considerably more elongated than the former segment, having three long swimming hairs and several spines at the distal margin. Genital area lying close to the posterior extremity of the ventral surface, immediately posterior to the epimeral region (Text-fig. 32, right). A fissure-like genital aperture is present, on both sides of which is present a pair of triangular genital plates, each bearing three acetabula and several hairs. Acetabula arranged in triangular form, the two anterior ones arranged side-by-side on the same level, the posterior one shifted slightly posteriorly to the anterior ones. Excretory pore opens at the posterior extremity of the dorsal surface, posterior to the dorsal shield. Colour and eyes as in the male.

Locality. One male and four females were collected from the main stream of the River Inôzawa, at Otiai, on June 8th, 1938.

Remarks. The present specimens show some significant features characteristic of *B. versicolor* (MÜLLER) commonly known in Europe and also reported from Siberia and Ussuri region. However, the facts that the epimeral region of the male of the present materials does not extend so far posteriorly as in *B. versicolor*, and that the genital aperture opens on the conspicuously protruded conical process, are likely to indicate slight deviation from *B. versicolor*. In view of these slight differences the writer has the intention to regard the present materials as a variety of the species mentioned above. However, a single male specimen having partially damaged legs cannot offer sufficient evidence to determine accurately the position of the present materials.

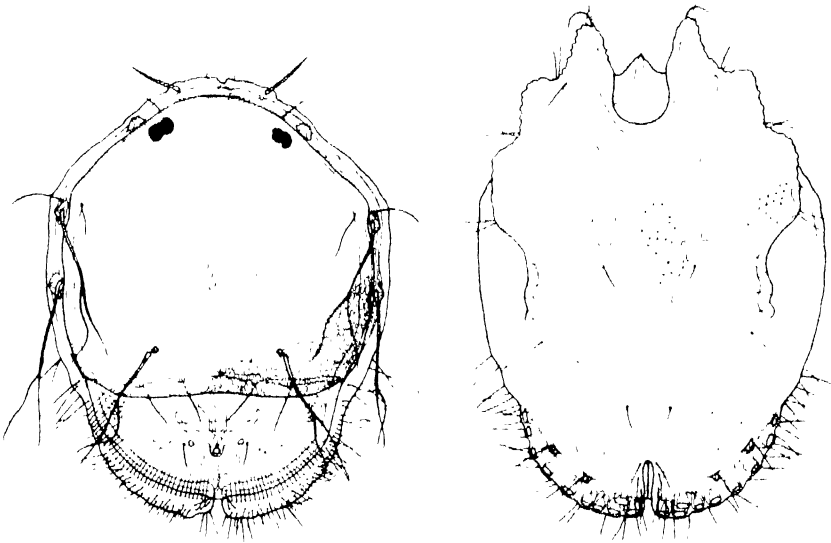


Text-fig. 33. *Brachypoda versicolor* MÜLLER, var.
Left, left palpus of female; right, right palpus of female.

10. *Aturus miyashitai* UCHIDA

(Text-figs 34-38)

Male. Body dorso-ventrally compressed, being of a thickness of about 0.16 mm. Length 0.30 mm; breadth 0.23 mm at the widest portion. General outline oval, tending to develop antero-lateral corners and showing somewhat extended loins, thus forming a pentagonal configuration with one angle protruding forwards (Text-fig. 34, left). A deep and moderately broad incision is present at the posterior end. A pair of



Text-fig. 34. *Aturus miyashitai* UCHIDA. Left, dorsal view of male; right, ventral view of male.

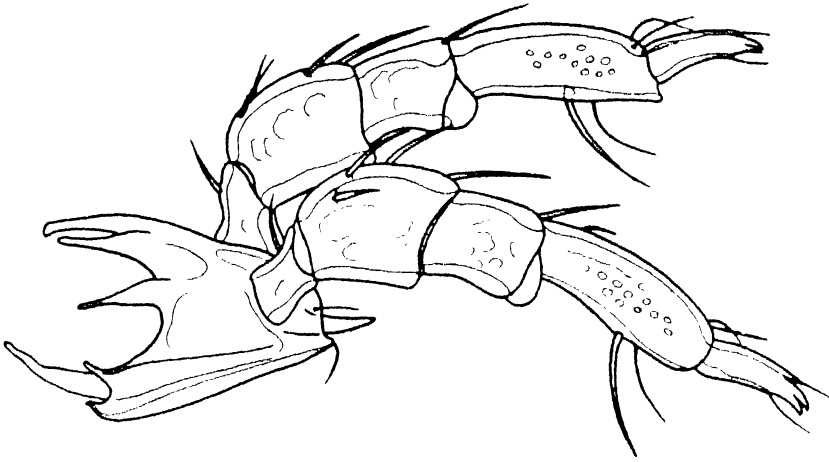
short and wide warts is found at the anterior extremity in a close position, each of these warts bearing a slender antenniform bristle near the outer side. Dorsal shield pentagonal in form, directing a gentle protrusion anteriorly, and terminating posteriorly in a truncated straight margin. The shield covering only the anterior two-thirds of the dorsal aspect of the body, being fitted to the anterior outline and slightly less in dimension than the dorsal aspect which is expressed by the dorsal extension of the ventral plate. Between the dorsal and the ventral plates is left a narrow stripe formed by dorsal groove with coarsely undulating striation. On the dorsal groove are found three pairs of large gland papillae, of which the most anterior pair is accompanied by a slender hair, and the two more posterior pairs by long bifurcated bristles. Such

two claws and having a few hairs.

Epimera united to form a broad ventral plate, I, II and III pairs of them being obvious in their outline owing to suture lines between the respective pairs (Text-fig. 34, right). First epimera projecting beyond the anterior margin of the dorsal aspect of the body, forming a wide pocket for the maxillary base between both-sided components. Second epimera also protruding beyond the anterior border of the body, the same being the case for the third epimera. Outer margin of these epimera ragged. Fourth epimera inconspicuous in their outline, being continuous to the posterior ventral plate. Epimera coarsely porose. Legs moderately long, stout, the length being increased in the more posterior ones. No swimming hairs present. Anterior three pairs shorter than the body length, but the fourth pair a little longer than the body length. Several spiny bristles arising around the distal margin and on the extensor surface of each segment, those at the distal flexor surface of segments II-III of the first and the second pairs being the longest. Fourth pair, stoutest of all, sexually characterized (Text-fig. 36). On the fourth segment one third of the distance from the distal end are arranged eight long whip-like bristles chiefly around the ventral surface of the segment, each attaining to a length exceeding the whole length of the fifth segment. Out of these are present several insignificant bristles and hairs on the dorsal surface, and along the extensor margin of the segment. At the distal extremity of the ventral surface and slightly deviated to the extensor side, a long scoop-like bristle protrudes which attains to almost the full length of the fifth segment. A slender spine is inserted at the same level with the scoop-like bristle at the distal extremity of the extensor margin. On the ventral proximal portion of the fifth segment are planted six bristles of moderate length. Distal segments of all the legs ending in two claws, each with two accessory claws and lamina. Genital area shifted to the posterior extremity of the ventral plate. Genital pore opening in the posterior fissure. Along the posterior margin are arranged about ten acetabula on both sides of the genital opening. A few protective hairs are accompanying this row of acetabula.

Colour brownish red anteriorly, and reddish purple at the posterior half. Eyes black.

Female. Body rounded, slightly shouldered, measuring 0.33 mm long and 0.27 mm wide at the widest portion. Body very much flattened, being 0.08 mm in thickness. Dorsal shield covering almost the whole dorsal surface, leaving a narrow stripe of dorsal groove around the outer

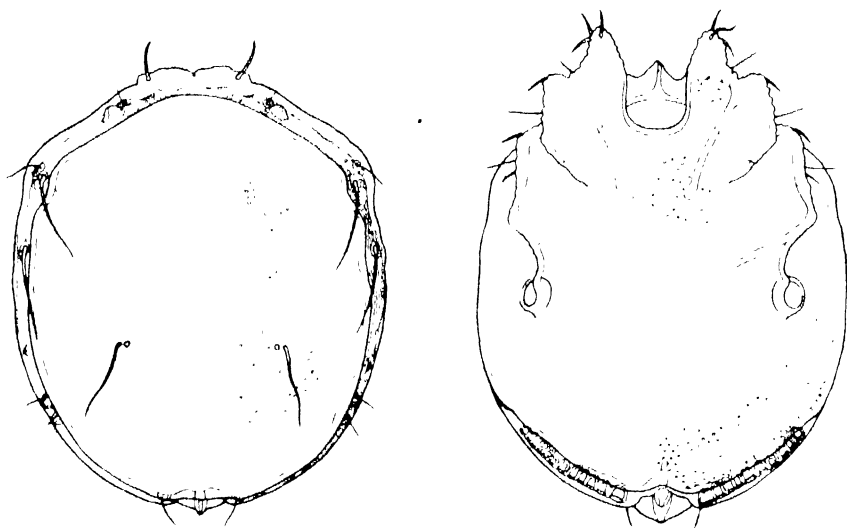


Text-fig. 37. *Aturus miyashitai* UCHIDA Palpi of female.

margin (Text-fig. 38, left). Texture of the dorsal shield is similar to that of the male. On the dorsal groove are found several pairs of gland papillae with or without accessory bristles or hairs, but no bifurcated long bristles detectable as is the case in the male. Excretory pore opens at the posterior end of the dorsal shield. Maxillary organ, mandibles and palpi showing close similarity to those of the male (Text-fig. 37). Epimera in general coinciding with those of the male. Legs much slenderer than those of the male, the three anterior pairs being a little shorter than the body length. Manner of arrangement of bristles is similar to that described in the male. Fourth pair of legs perceptibly longer than the body, being quite different from the fourth pair of the male. Fourth segment slender, being simply provided with a few marginal bristles and a few spines on the flexor margin. Fifth segment exactly similar to the fourth. Genital opening at the posterior extremity. About ten or more acetabula arranged in a row along the posterior margin of the ventral plate (Text-fig. 38, right). Colour reddish brown anteriorly, and purple at the posterior half. Eyes black.

Locality. One male and one female were collected together with the following species (*Aturus caudatus*, n. sp.) from the main stream of the River Inôzawa, at O'Kiti-ga-Futi, on June 8th, 1938.

Remarks. The species is also reported by UCHIDA (1934) on the basis of materials collected from the River Yura. The present specimens show several important characteristics which agree well with UCHIDA's description, although they differ slightly in some insignificant points.



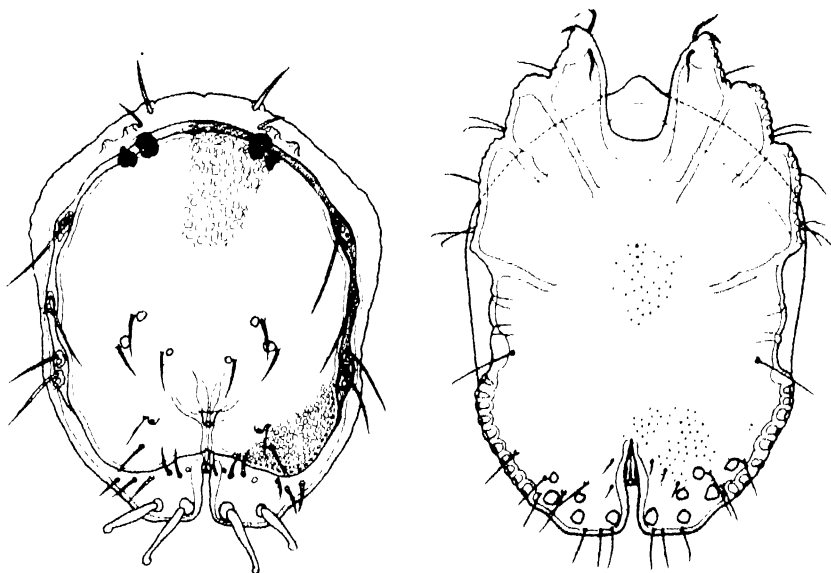
Text-fig. 38. *Aturus miyashitai* UCHIDA. Left, dorsal view of female, right, ventral view of female.

11. *Aturus caudatus*, n. sp.

(Text-figs 39-43)

Male. Body rather angular at some five corners, extending anteriorly a well-arched frontal portion and remarkable shoulders. Length attaining to 0.28 mm and breadth to 0.22 mm at antero-lateral corners. Body flat, being about 0.11 mm in thickness. Most of the dorsal surface covered by a wide dorsal shield which has somewhat pentagonal outline, well fitted to the free margin of the body, but is considerably smaller in area at antero-lateral and posterior regions (Text-fig. 39, left). Surface of the dorsal shield is universally furnished with coarse pores, the texture exhibiting a sort of reticular appearance of irregular polygonal forms under low power magnifications. Four pairs of gland papillae with accompanying protective bristles and a pair of bristles are found on the posterior half of the dorsal shield. Around the margin of the dorsal shield, from the anterior extremity to the postero-lateral sides, is present a narrow band formed by a dorsal groove, outside which extends the dorsal extension of the ventral plate forming the general contour of the dorsal aspect of the body. Along the dorsal groove are present four pairs of gland papillae, each accompanied by a simple elongated bristle. Eyes double, situated on the dorsal groove near the anterior end. Directly

outside the eyes are observed two pairs of gland papillae, the anterior pair being provided with accessory bristles. Antenniform bristles stout, not very long, inserted at the outer margin of a pair of insignificant broad elevations. Posterior one sixth of dorsum uncovered by the dorsal shield, the uncovered portion being the dorsal extension of the ventral plate. Surface of this area smooth. This area is divided into right and



Text-fig. 39. *Aturus caudatus*, n. sp. Left, dorsal view of male; right, ventral view of male.

left regions by a deep narrow posterior incision. Along the posterior border of the dorsal shield are arranged several minute bristles in a row. Distinct from this row of short bristles, and close to the posterior truncated edges of the body, are found four long club-shaped bristles which terminate in rounded heads and which are inserted respectively in four large pores. These two pairs of the club-shaped bristles are very impressive even at a glance. Maxillary organ 0.07 mm long, rather thick, and about one half of length of palpi (Text-fig. 40, c). Mandibles a little longer than the maxillary organ. Palpi (Text-fig. 40, a & b) porose, of elegant form, overlapping one half of body length. Lengths of each palpal segment are (in mm):

	I	II	III	IV	V
Extensor side.....	0.02	0.06	0.02	0.06	0.03
Flexor side.....	0.02	0.03	0.02	0.05	0.03

First segment short and slender, with one minute bristle on the extensor edge. Second segment, broadest of all, with gently convexed extensor side and perceptibly concave flexor side. Three moderate-sized bristles are arranged along the extensor margin. At the distal extremity of the flexor surface a blunt triangular process protrudes from the inner

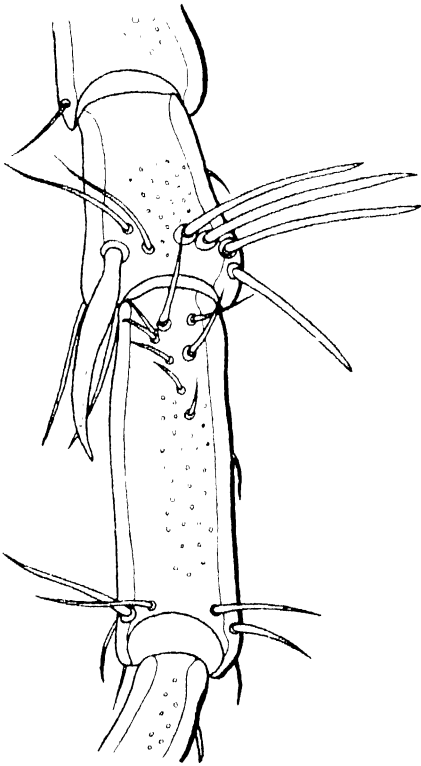


Text-fig. 40. *Aturus caudatus*, n. sp. a, right palpus of male; b, left palpus of male; c, side view of maxillary organ with mandible.

side of the segment. Third segment narrower than the second, slightly curved at the flexor surface. Extensor side is provided with a distal minute bristle. Fourth segment club-shaped, tapering in slight degree towards proximal and distal directions. Flexor surface obviously convexed at the midway point between the two extremities. A short bristle and a long hair are planted at the neighbourhood of this convexity. At the distal end of the extensor side only one hair is present. Fifth segment ending in two claws, being provided with a few hairs.

Epimera united to form a broad plate covering more than anterior half of venter. The anterior three pairs of epimera are respectively extending their anterior processes beyond the anterior margin of the body (Text-fig. 39, right). Sutures between the I, II and III epimera are obvious, but those between the IV epimera and the posterior ventral

plate vanishing. Third epimera very wide, occupying almost the total area of I plus II epimera. Outer margin of all the epimera distinctly ragged. Legs robust, wholly destitute of swimming hairs, increasing in length posteriorly. First and second pairs scarcely comparable with the body length, but third and fourth pairs exceeding the body length. Around the distal margin and on the dorsal side of each segment are stout spines sparsely arranged. Segments II and III of the first and second pairs



Text-fig. 41. *Aturus caudatus*, n. sp. III-VI segments of right fourth leg of male, ventral view.

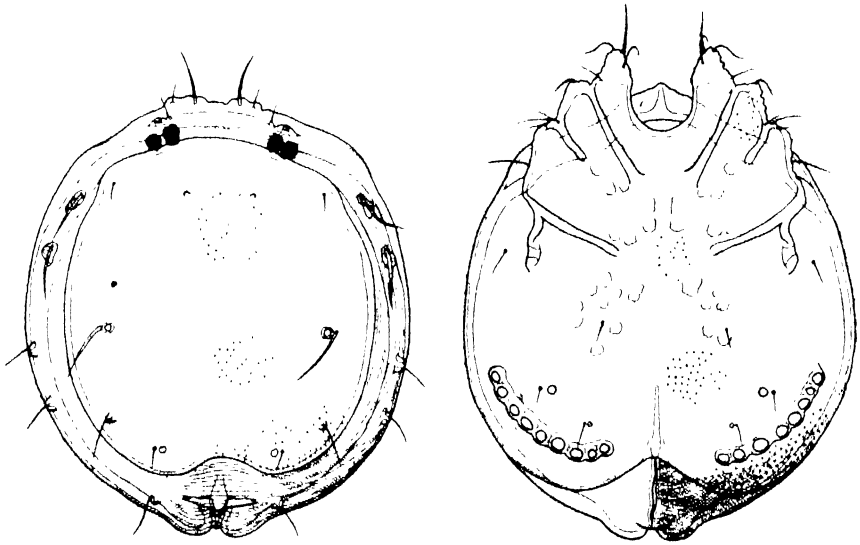
have a few long bristles at the distal end of the flexor side. Fourth pair has only a few minute spines and hairs, except on the fourth and the fifth segments, these two being sexually modified (Text-fig. 41). Fourth segment very much reduced in length as compared with those in the other pairs of legs, being provided with one large strong horn-shaped spine at the distal end of the ventral surface. This spine is about one half the length of the fifth segment. Around this specialized spine are arranged several long bristles, of which the four arising from the flexor region are long and stout. Dorsal surface of the fourth segment devoid of any significant spines or bristles. Fifth segment elongated, with about nine minute bristles on the ventral surface of the proximal region and with several

longer bristles around the distal margin of the segment. Sixth segment of all legs ending in two claws, each with three accessory claws and narrow lamina. Excretory pore opens on the dorsal side near the posterior margin of the dorsal shield. Genital opening in the posterior fissure. Along the postero-lateral margins are arranged about fifteen acetabula roughly in a row. Of these acetabula the posterior five or six

have wide external openings. Several slender hairs protecting the genital region.

Colour brownish red. Eyes black.

Female. Body rounded in outline, slightly shouldered at antero-lateral corners. Length 0.38 mm, width 0.33 mm at the widest middle portion. Thickness ca. 0.15 mm, considerably compressed dorso-ventrally. Dorsal shield occupying the wide central portion of the dorsal aspect of the body, being of almost similar in outline to the general contour of dorsum except for an inward curvature at the posterior end (Text-fig. 42, left).



Text-fig. 42. *Aturus caudatus*, n. sp. Left, dorsal view of female, right, ventral view of female.

Sculpture of the shield resembling that of the male. Three pairs of gland apertures with accessory bristles or hairs are found on the posterior half of the plate. Eyes in the dorsal groove which encircles the dorsal shield with extended breadth. Four pairs of gland papillae and accessory hairs arranged along the lateral margin of the body on the dorsal groove. Posterior part of dorsum is occupied by coarsely striated skin which is continued from the dorsal groove. Maxillary organ, mandibles and palpi resemble those of the male (Text-fig. 43). Second segment of palpus is provided with several bristles on the extensor surface, much more numerous than in the male, and on the extensor edge of the fourth segment is found a minute spine approximately at the midway point between the

two extremities of the segment. Epimera similar to those in the male (Text-fig. 42, right). Legs much slenderer than those of the male. Characteristics of the arrangement of bristles and spines agree with those



Text-fig. 43. *Aturus caudatus*, n. sp. Palpi of female.

in the male. Fourth segment of the fourth pair elongated, showing no specialized arrangement of special spines or of bristles. Ventral plate does not reach the posterior extremity, but is replaced by non-chitinous skin which is divided into right and left parts by a fissure-like genital aperture. Excretory opening on the dorsal surface, at the posterior extremity. Along the posterior border of the ventral plate, and a little apart from the latter, are arranged about nine acetabula in a row. Two pairs of hairs protecting this area. Colour and eyes same as the male.

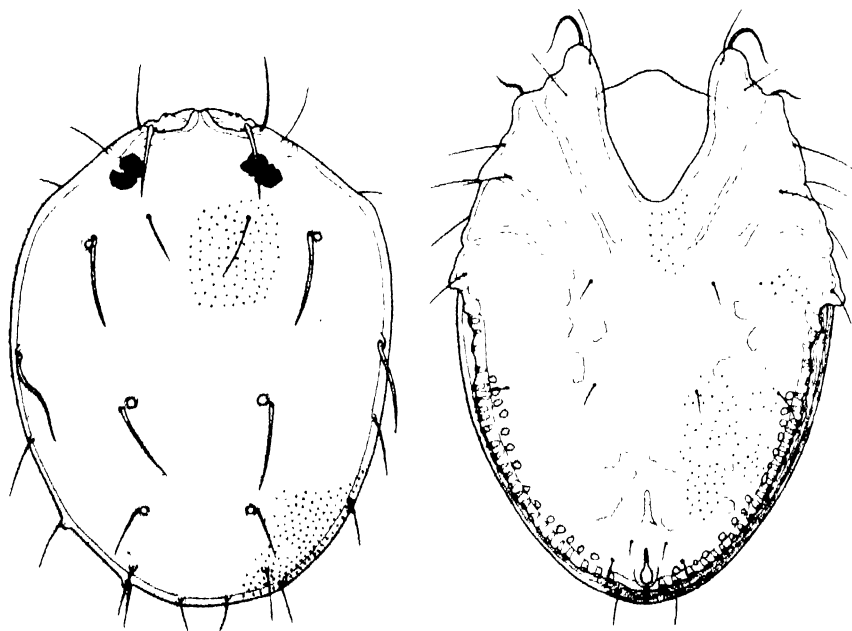
Locality. One male and one female were secured at O'Kiti-ga-Futi, from the River Inôzawa, on June 8th, 1938.

Remarks. The writer has been unable, in intensive references to scientific works, to find mention of any species allied to the species under discussion. The four posterior appendages of club-shaped chitinous spines and the arrangement of specialized bristles on the fourth segment of the fourth leg of male appear to characterize uniquely the present specimens.

12. *Kongsbergia materna* THOR

(Text-figs. 44-46)

Male. Body inverted oval, minute, measuring 0.28 mm long and 0.19 mm wide at the widest portion. Seen from the side the body is fairly flattened, being of 0.08 mm of thickness. Dorsal shield hard, extending posteriorly beyond the extremity of the ventral plate (Text-fig. 44, left). The latter, however, extending anteriorly beyond the dorsal shield. Chitinous plates of dorsum and ventor are universally sculptured with coarse pores. Groove between the dorsal and the ventral plates is found on the body-sides in the anterior part, and is shifted to the ventral margin in the posterior part. Goove is rather soft-skinned, containing several hairs and bristles along its course. Eyes double, situated near the an-

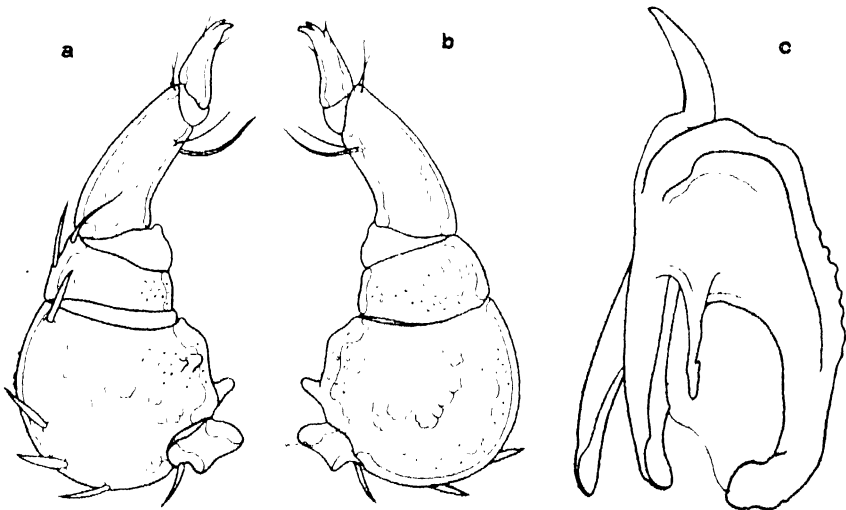


Text-fig. 44. *Kongsbergia materna* THOR. Left, dorsal view of male; right, ventral view of male.

terior margin of the dorsal shield, both-sided ones being in very close proximity. Antenniform bristles slender and elongated. On the dorsal shield are present two rows of gland papillae and accessory hairs. Maxillary organ large in relation to the dimensions of the body, and occupies more than one third of the latter (Text-fig. 45, c). Mandibles 0.14 mm long, surpassing the length of the maxillary organ by the length of claws. Palpi (Text-fig. 45, a & b) exceedingly large as compared with the body, attaining to far more than one half of the body. Their appearance is very thick and stout: the lengths of palpal segments are (in mm):

	I	II	III	IV	V
Extensor side.	0.02	0.15	0.04	0.08	0.04
Flexor side.	0.03	0.06	0.03	0.05	0.04

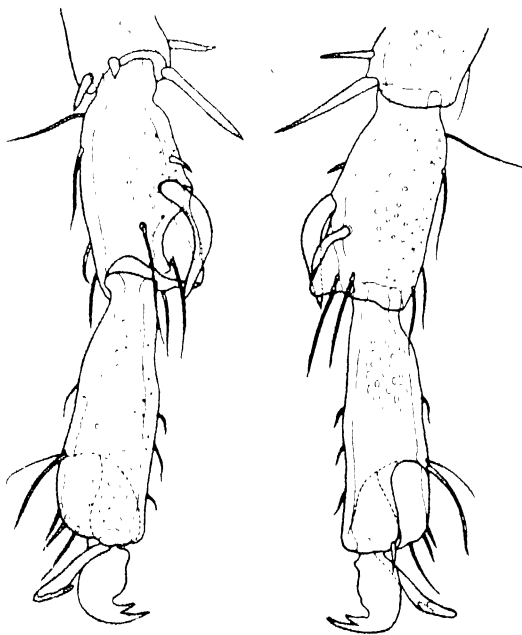
Of all the segments the second is the broadest, with well-arched semi-circular extensor surface and remarkably convexed flexor margin. Along the extensor margin are inserted four stout bristles, one distal and three proximal in position. Near the proximal end of the flexor surface there is a short tubercle of papilla-shape, and distally, a little distal from the middle region, are seen two small conical papillae on the inner surface. Distal part of the flexor surface is sculptured with coarse



Text-fig. 45. *Kongsbergia materna* THOR. a, left palpus of male, inner side; b, left palpus of male, outer side; side view of maxillary organ with mandibles.

pores. Third segment short, but broad, having two bristles of moderate length on the extensor margin. Fourth segment long, obviously narrowed towards the distal end. Bristles and hairs on this segment are very sparse, being represented by only a long hair and a bristle near the distal extremity of the flexor surface, and by a few minute hairs at the distal end of the extensor surface.

Epimera united to form a continuous ventral plate (Text-fig. 44, right). First pair are oblique narrow stripes, protruding beyond the anterior body margin, embracing a long and wide pocket for the maxillary base. Second pair resembling the first one. Third pair small and narrowed towards the medial portion, leaving well-arched suture lines between the third and the fourth epimera. Fourth pair indistinguishable from the remaining portion of the ventral plate, since no visible suture line is found between these two regions. Legs short and robust, devoid of swimming hairs, but several stout bristles are found around the distal margins of the segments except around the Vth, which ends in two small claws. The Vth segment of the fourth leg is sexually characterized (Text-fig. 16). Distal half of the flexor margin is significantly concave; at the proximal end of this concavity are present one large horn-shaped spine and one small accessory spine, the latter being inserted dorsally to, and slightly distally to the former. Out of these spines several long hairs are growing encircling the distal extremity of the segment in question. Genital opening lying accessory to the posterior end of the ventral plate. On both sides of the genital opening, and along the free margin of the ventral plate, are arranged numerous minute acetabula in two rows. Ex-



Text-fig. 46. *Kongsbergia materna* THOR. Left, IV-VI segments of right fourth leg of male, ventral view; right, the same, dorsal view.

cretory pore opens ventrally, immediately posterior to the genital opening.

Colour brownish red. Eyes black.

Locality. Three males were collected from the main stream of the River Inôzawa, at Mitukuri, on June 8th, 1938.

Remarks. This is the first report of the Genus *Kongsbergia* from Japan. The present specimens show a marked resemblance to *K. materna* THOR, a common European species distributed in various regions of that continent. Although a slight difference is detectable in the size of the second palpal segment, and in the position of the distal bristle at the flexor surface of the fourth palpal segment, the writer is of the opinion that the present materials should be identified with *K. materna* mentioned above, regarding such disagreements to be local variation only.

LITERATURE CONSULTED

- LUNDBLAD, O. 1927. Die Hydracarina Schwedens. I. Beitrag zur Systematik, Embryologie, Ökologie und Verbreitungsgeschichte der Schwedischen Arten. Zool. Bid. Uppsala, Bd. 11.
- LUNDBLAD, O. 1929. Wassermilben aus dem nordöstlichen Altai. Entomol. Tidskr., Ht. 3-4, 1929.
- LUNDBLAD, O. 1930. Hydracarina. Zoology of the Faroes. Copenhagen.
- LUNDBLAD, O. 1934. Report on Hydracarina. Mem. Conn. Acad., Vol. 10, Art. 7.
- LUNDBLAD, O. 1936. Über einige Hydracarina aus Java. Entomol. Tidskr., Ht. 2-3, 1936.
- MARSHALL, R. 1932. Preliminary List of the Hydracarina of Wisconsin. Part II. Trans. Wisconsin Acad. Sci. Art. & Lett., Vol. 27.
- MARSHALL, R. 1933. Water Mites from Wyoming as Fish Food. Trans. Amer. Micr. Soc., Vol. 52.
- MONTI, R. 1910. Contributo alla biologia degli Idracnidi Alpini in relazione all'Ambiente. Att. Soc. Italiana Sci. Nat., Vol. 49.
- MOTAS, C. 1928. Contribution à la connaissance des Hydracariens français, particulièrement du Sud-Est de la France. Trav. Lab. d'Hydrobiol. et Piscicult. Univ. Grenoble, 20.
- MOTAS, C. 1933. *Aturus (Crinaturus) spatulifer* PTERSIG in den rumänischen Karpathen. Zool. Anz., Bd. 103.
- SOKOLOW, I. 1930. Die Hydracarina von Russisch-Karelien. Zool. Jahrb. Syst., Bd. 59.
- SOKOLOW, I. 1930. Beiträge zur Kenntnis der Hydracarina Sibiriens. Arch. Hydrobiol., Bd. 22.
- SOKOLOW, I. 1934. Beiträge zur Kenntnis der Hydracarinafauna des Ussuri-Gebietes. II. Hydracarina der fließenden Gewässer. Zool. Jahrb. Syst., Bd. 65.
- SZALAY, L. 1933. Über zwei Wassermilbenarten aus der Gattung *Atractides* C. L. KOCH. Zool. Anz., Bd. 102.
- SZALAY, L. 1933. Zwei neue Wassermilben aus der Gattung *Atractides* C. L. KOCH. Zool. Anz., Bd. 103.
- SZALAY, L. 1933. Eine neue Hydracarina aus der Gattung *Atractides* C. L. KOCH und das Weibchen von *Atractides (R.) ungeri* SZALAY. Zool. Anz., Bd. 104.

- SZALAY, L. 1935. Zwei neue Hydracarinae aus der Gattung *Megapus* NEUMAN und das Weibchen von *Megapus* (*M.*) *barsiensis* SZALAY. Zool. Anz., Bd. 110.
- SZALAY, L. 1935. Eine neue Hydracarinae aus der Gattung *Megapus* NEUMAN und das Weibchen von *Megapus* (*M.*) *nodipalpis* var. *fluvialis* SZALAY und *Megapus* (*M.*) *acutirostris* MOTAS. Zool. Anz., Bd. 111.
- UCHIDA, T. 1931. Einige Wassermilben aus Japan. Zool. Anz., Bd. 95.
- UCHIDA, T. 1934. Some Rheophilous Water-mites from Japan. J. Fac. Sci. Hokkaido Imp. Univ., Zool., Vol. 3.
- UCHIDA, T. 1936. Water Mites from the Kurile Islands. Bull. Biogeogr. Soc. Japan, Vol. 6.
- VIETS, K. 1935. Die Wassermilben von Sumatra, Java und Bali nach den Ergebnissen der Deutschen Limnologischen Sunda-Expedition. Arch. Hydrobiol., Suppl.-Bd. 13-14.
- VIETS, K. 1936. Die Tierwelt Deutschlands usw. 31, 32 Teil. VII: Wassermilben oder Hydracarina. Jena.
- WALTER, C. 1928. Zur Kenntnis der Mikrofauna von Britisch Indien. Rec. Indian Mus., Vol. 30.
- WALTER, C. 1935. Hydracarina. Voyage de Ch. Alluaud et P. A. Chappuis en Afrique occidentale française (Dec. 1930-Mars 1931). Arch. Hydrobiol., Bd. 28.
- WILLIAMSON, W. and C. SOAR, 1915. British Hydracarina: The genus *Lebertia*. J. Quekett. Micr. Club, Vol. 12.

TERRESTRIAL OLIGOCHAETA FROM MANCHOUKUO

By

SHINJIRÔ KOBAYASHI

Keiyô Second Higher Common School

(With 15 Text-figures)

(Received March 11, 1940)

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INTRODUCTION

No earthworms having ever been reported upon from either Manchoukuo or Mongolia*, a survey of them from the zoogeographical point of view in these two countries had been on my part a long-cherished desire. Having obtained the financial aid from the Chosen Academy of Natural Science, I had at length the opportunity to study the terrestrial oligochaete fauna of Manchoukuo, during months of July and August, in 1937. The present report is, of course, only a preliminary one. But, as I consider that the number of species will not be markedly increased by any further researches, I intend to publish here the outline of the terrestrial oligochaete fauna of Manchoukuo.

As the greater part of Manchoukuo is of a continental climate, it is not likely that an abundance of earthworm-fauna is to be found there. But, the present survey has at least made clear some important zoogeographical and ecological facts. Namely, (1) the rainfall and the temperature are the most important factors in delimiting the distribution of the terrestrial oligochaetes; (2) the northern boundaries of both genera *Pheretima* and *Drawida* are to be drawn through Manchoukuo; (3) the southern part of this country is important in considering the distribution of the family Lumbricidae.

ACKNOWLEDGMENTS

I am greatly indebted to the Chosen Academy of Natural Science, which made it possible for me to carry out the study by allowing me a generous grant. I also wish to express my thanks to the authorities of the Administrative Office of Manchoukuo, to the Korean Office of Man-

* MICHAELSEN ('01) reported the occurrence of *Bimastus beddardi* in N.-E. Mongolia. But, recently ('31) he himself stated that no oligochaetes are known from Mongolia.

After the completion of this manuscript, *Eisenia foetida* was reported by MISAHA ('38) from Jehol, Manchoukuo.

choukuo, and to the Bureau of School Affairs of the South Manchurian Railway Company, for their assistance in many ways throughout the survey. I should like to offer my cordial thanks to Dr. SANJI HÔZAWA, Professor of the Tôhoku Imperial University, for his continual guidance during the course of my study. I also wish to express my hearty thanks to the following gentlemen, who kindly sent me the materials taken from various localities in which I was not able to make collections myself; Mr. SUKENDO KOHASI and Mr. HIROSI TADA (Chihfen, Jehol), Mr. KAZUO KOBAYASHI (Fengtien and its vicinity), Mr. JUTTI ÔIZUMI (various localities in the Kwanto Province, and Wangyehmiao), and Mr. KUNIO SUDA (Anshan and its vicinity). I also wish to express my cordial thanks to Mr. SINKEI GEN of the Forest Experiment Station, General Government of Chosen, for his kindness shown in determining pH of the soil of the habitats. Furthermore, I must pay my sincere thanks to the members of the two offices, the Satumen Konsu at Ilikete and the Paichengtze Office of the S.M.R.C. at Halun-Arshan, who gave me invaluable assistance during my stay in those places.

Systematics

Family Moniligastridae

Genus *DRAWIDA* MICHAELSEN 1900

1. *Drawida japonica* (MICHAELSEN) 1892

1938 *Drawida japonica*, KOBAYASHI, Sci. Rep. Tôhoku Imp. Univ., Biol., XIII, 2, pp. 94-95, fig. 1.

According to GATES ('35), *Dr. propatula* is distinguishable from its most closely allied species *Dr. japonica*, by its larger size ($113-130 \times 4$ mm in contrast with $28-65 \times 1-2$ mm) and by the absence of the elongate rod-like appendices on the ovisacs. The same writer's remarks on *Dr. grahami* ('35) is as follows "Distinguished from *Dr. japonica* by the absence of the appendix on the ovisacs, . . .". CHEN has written in his recent paper ('36) that *Dr. grahami* is synonymous with *Dr. japonica* and that the appendix on the ovisac has no systematic value. Recently ('38), I have expressed my opinion after studying the Korean material that the posterior appendix of the ovisac may have systematic value.

In the present investigation, as I met with examples of both *Dr. japonica* and *Dr. propatula*, I was able to examine the systematic value of the posterior appendix of the ovisac. Of *Dr. japonica* seventy-six mature specimens (together with several Korean specimens) were dissected

and of *Dr. propatula* 21 mature specimens were dissected as well as several immature ones of both species.

In the case of *Dr. japonica*, an ovisac consists of three parts of (a) anterior part, (b) posterior part, and (c) extreme terminal end. (a) The anterior part extends interior of three segments of XIV-XVI, lying in the dorsolateral side of the alimentary tract, and is filled up with ripe ova and is moniliform in form being constricted by the septa. (b) The posterior part is continuous with the part (a). It is always thinner than (a), and is free from the septa or weakly constricted by them, its interior either filled up or partially filled with ripe ova. Of this posterior part we may distinguish the following two types. (i) It is short and twisted running along the dorsolateral or lateral side of the alimentary tract, and is free from the septa. (ii) It runs straightly caudalwards underneath the alimentary tract, and is flattened being compressed by the gut and is weakly constricted segmentally. In the case of (ii) it is not rare that it extends posteriorly beyond XXX (even in the immature specimens); while in the case of (i) it is always shorter than in (ii), and is sometimes only of 3-5 segmental length. More than half of the examined specimen (42/76) were of the type (ii). But, in the case of two specimens the intermediate type was found; i. e. in one specimen, one side was of type (i) while the other side was type (ii). This posterior part of the type (ii) may be called "posterior rod-like appendix". Its unusual appearance surely attracts our notice when observed, though it has been overlooked by MICHAELSEN and STEPHENSON as GATES pointed out ('35). Apparently, the correlation existing between the locality and the high frequency of the occurrence of this organ was not clearly recognizable. (c) The extreme terminal end of the ovisac which is thin and is only of 1-2 segmental length, appears usually empty and appears whitish in colour. This part may be easily overlooked.

In the case of *Dr. propatula*, an ovisac extends interior of the segments of XVI-XIX. As in the case of *Dr. japonica*, it consists also of three parts of (a), (b) and (c). (a) The anterior part situated on the dorsolateral side of the alimentary tract, is moniliform in form being constricted in various degrees by the septa on its course and its interior is fully filled with ripe ova. (b) The posterior part of 2-5 segmental length, is a little thinner than (a), and is usually slightly twisted on the dorsolateral or lateral side of the alimentary tract, and is also fully filled with ripe ova. (c) The extreme terminal end which is only of 1-2 segmental length, appears always empty and is much thinner than (b); this part

may be easily overlooked. Thus, when the part (a) is weakly constricted by the septa, the ovisac appears finger-like.

As mentioned above, at least in all the examined specimens, the "posterior rod-like appendix" was not found in the case of *Dr. propatula*. In *Dr. japonica*, out of 76 specimens examined, 42 were provided with this appendix; among the material collected from every locality, at least a few specimens were found to have this characteristic organ; sometimes, most of the specimens collected in one locality were found to have this organ. From this result, it may be easily judged that, although the "posterior appendix" in the case of *Dr. japonica* may be, of course, a unique characteristic, but it is systematically not so important as GATES has thought. Thus, this organ may not be taken as a criterion for *Dr. japonica*, but may be utilized as a supplementary mean.

Body length 34–81 mm, greatest diameter 2.2–3.6 mm. The dimensions 28–65 × 1–2 mm which were given by GATES ('35 a) of the body size of the present species, seems to be smaller than that of the Manchoukuo specimens (and also of the Korean specimens).

Localities and materials: Harbin, 6 mature and 2 immature sps.; Kirin, 34 mature and 109 immature sps.; Tumen, 4 mature and 21 immature sps.; Paichengtze (=Taoan), 14 mature and 11 immature sps.; Fengtien, 25 mature and 14 immature sps.; Fsfeng, 1 mature sp.; Tashihchiao, 11 mature and 11 immature sps.; Chihnsien, 36 mature and 5 immature sps.; Antung, 8 mature sps.

Distribution: Japan, Korea, Manchoukuo, China, India.

2. *Drawida propatula* GATES 1935

1933 *Drawida japonica* (part), CHEN, Contr. Biol. Lab. Sci. Soc. China, Zool. Ser., IX, 6, pp. 189–194, fig. 3.

1935 *Drawida propatula*, GATES, Lingnan Sci. Jour., XIV, 3, pp. 449–450.

Description:

External characteristics:

Length 73–114 mm, greatest diameter 4.0–5.0 mm; largest specimen 114 × 4.8 mm and the smallest 74 × 4.0 mm; usual size 95 × 4.5 mm. Number of segments 149–179. Colour in formalin, unpigmented or uniformly light blue due to the content of the alimentary tract; clitellum reddish. Clitellum well-marked, moderately swollen, extending from IX to a part (1/3–1/2) of XIV.

Setae small, closely paired, beginning on II; cd on X– or XI–XIII

smaller than the rest. Setal distance $ab=cd$; on the preclitellar segments, aa is very slightly greater than, or subequal to, bc ; on the postclitellar segments, aa is slightly smaller than bc ; in one specimen picked up at random, $aa:bc=33.5:32$ preclitellarly, $45:58$ immediately posteriorly to the clitellum, $39:53$ in the middle portion of the body; dd is slightly greater than, or subequal to, $1/2$ of the circumference.

Male pores are minute, transverse slits on X, nearer to b than to c , and nearer to the intersegmental furrow 10/11 than to the transverse setal line; each on a very small, oval or circular, not sharply demarcated, whitish porophore which is situated on a small, indistinctly demarcated, oval epidermal elevation. The anterior border of this elevation does not reach the posterior one third of X, and the posterior margin is just on the intersegmental furrow 10/11 which is slightly displaced posteriorwards. In general, its appearance is very similar to that of *Dr. japonica*.

Female pores are scarcely visible on the anteriormost edge of XII, on ab -line.

Spermathecal pores are found within crescent-shaped depressions placed at the posterior margin of VII, just medial to c , or sometimes between b and c but much nearer to c ; each pore represented as a minute, longitudinal, diagonal or transverse slit. Usually a very small spermathecal papilla is found within this slit.

Genital papillae are similar in shape to those of *Dr. japonica*. They are found on VII–XI; more frequently on VIII–X, less frequently on XI, and rather rarely on VII; 1–8 in number, and in most cases 3–5; frequently those on X are pairly occurred though they are not always strictly symmetrical in position; in most cases, that on XI is single and midventral; those on the other segments are considerably variable in position.

Internal anatomy:

Septa 5/6–8/9 much thickened, and the remaining ones thin or membranous; 9/10 and 12/13 slightly displaced posteriorly, dorsal part of 10/11 displaced to the anterior part of XII, the others almost normally inserted.

In all the examined 21 specimens, gizzards are two, in XII–XIII.

Testis sacs constricted by 9/10, usually the larger part lying in IX, about 1.9–2.5 mm in length and 1.3–2.0 mm in width. Sperm-duct is short with a few loose loops and passes into the parietes just antero-medially to prostate, about 5–7 mm long. Prostate large, thumb-shaped, usually slightly compressed at about ectal third, moderately warty on

surface, about 1.8–2.6 mm long and 0.8–1.2 mm wide. Central body slenderly tubular.

Ovarian chamber closed off dorsally, but attached to the parietes laterally, widely separated ventrally. Ovisac moniliform or finger-shaped, extending posteriorly into XVI–XIX, not provided with posterior appendix.

Spermatheca with atrium lying behind $7/8$; ampulla spherical, about 0.8–1.3 mm in diameter; spermathecal duct long, about 8–10 mm long, looped, not sharply marked off from the ampulla, ectally passing into the lateral side of atrium in the middle portion. Atrium columnar, about 1.0–1.8 mm high, usually a little or a very little longer than the diameter of ampulla.

Accessory glands tough-walled, sessile but moderately protruded into coelom.

Locality and material: Yenki, 10 mature, 51 immature and juvenile sps.

Distribution: Central China (Kiu-Kiang, Kiangsi), Manchoukuo.

Remarks:

The body, prostate and spermathecal duct of the Manchoukuo specimens are slightly smaller than those in GATES' material, but are distinctly larger than those of *Dr. japonica*. Further, I must mention here that all the present specimens are extremely contracted. The ovisacs have no posterior rod-like appendices as already mentioned as being the feature of *Dr. japonica*, but the other characteristics are similar to those of as described of this species.

Generally, the body-size is one of the most variable characteristics. The present species has been obtained from only two localities in China and Manchoukuo; and as the specimens examined by GATES and by myself are rather few in number, the extent of the variation of the body-size is not yet clearly determined though it may possibly be greater than that of *Dr. japonica*. Furthermore we must remember that the body-size of both the Korean and Manchoukuo specimens of *Dr. japonica* is larger than that of the Chinese specimens of the same species. The presence or absence of the posterior appendix in the ovisac seems to have no systematic value.

Although the present species appears to be a distinct species, a further examination will be needed to establish the validity of this assumption.

3. *Drawida koreana* KOBAYASHI 1938

1938 *Drawida koreana*, KOBAYASHI, Sci. Rep. Tōhoku Imp. Univ., Biol., XIII, 2, pp. 102-107, fig. 3.

Male porophore is of characteristic as mentioned in the case of original specimens, being nipple-like or rather conical in shape. Spermathecal duct is short, thick and weakly muscled, and is about 3 mm long. Even in the immature specimens, the accessory glands of genital papillae are, however, poorly developed. In all the opened specimens, gizzards are three in number and located in XII-XIV. Unfortunately, as the specimens have been all macerated, the further examination was not able to make.

Localities and materials: Anshan and its vicinity, 14 mature and 7 immature sps., August, '38, by Mr. KUNIO SUDA; Fengtien, a single immature sp., August 15, '37, by Mr. KAZUO KOBAYASHI.

Distribution: Central and North Korea, Manchoukuo.

4. *Drawida jeholensis*, n. sp.

(Fig. 1)

Description:

External characteristics:

Length 52-66 mm, greatest diameter 2.8-3.5 mm, number of segments 153-160. The region of the preclitellar segments is somewhat conical in shape, and resembles that of *Dr. koreana* and of *Dr. nemora*. Colour in formalin, uniformly whitish grey.

Clitellum in IX-XIV, slightly swollen and slightly glandulated: glandularity of XIV is less developed. In most of the specimens, the ventrolateral parts of both segments X and XI are also slightly glandulated and projects more or less ventralwards.

Dorsal pores absent; epidermis along the middorsal line is not thin.

Prostomium prolobous.

Setae beginning on II, closely paired, small and not conspicuously projected, apparently a little stouter anteriorly than posteriorly. Setal distance ab is equal to cd ; aa is slightly larger than bc in the preclitellar region, but is smaller than the latter behind the clitellum; dd is subequal to, or slightly smaller than, $1/2$ of the circumference. Setal distances taken from a specimen are as follows: $aa:ab:bc:cd:dd=17.2:1.2:14.5:1.2:74.5$ on a preclitellar segment, $24:6:1.4:27:1.4:102.6$

on a segment immediately posterior to the clitellum, 21:1.6:28:1.5:79.7 in middle portion of the body.

Male pores are minute, in the form of transverse slits, seated on the top of somewhat conical or mamma-like porophore with oval base; the base of the porophore is about 0.7-0.8 mm in diameter and occupies about posterior $1/3$ - $2/5$ of X. The posterior margin of the porophore is always placed on the anteriormost part of XI beyond the intersegmental furrow 10/11, and the pore is found in the position corresponding to 10/11, between b and c but nearer to b. No trace of penis is found. In the anterior side to the male porophores, the posterior furrow of the middle annulus of X (X with 3 annuli) is so deeply grooved, that it appears to be an intersegmental furrow (Fig. 1, a).

Female pores are also in the form of transverse slits, on anteriormost

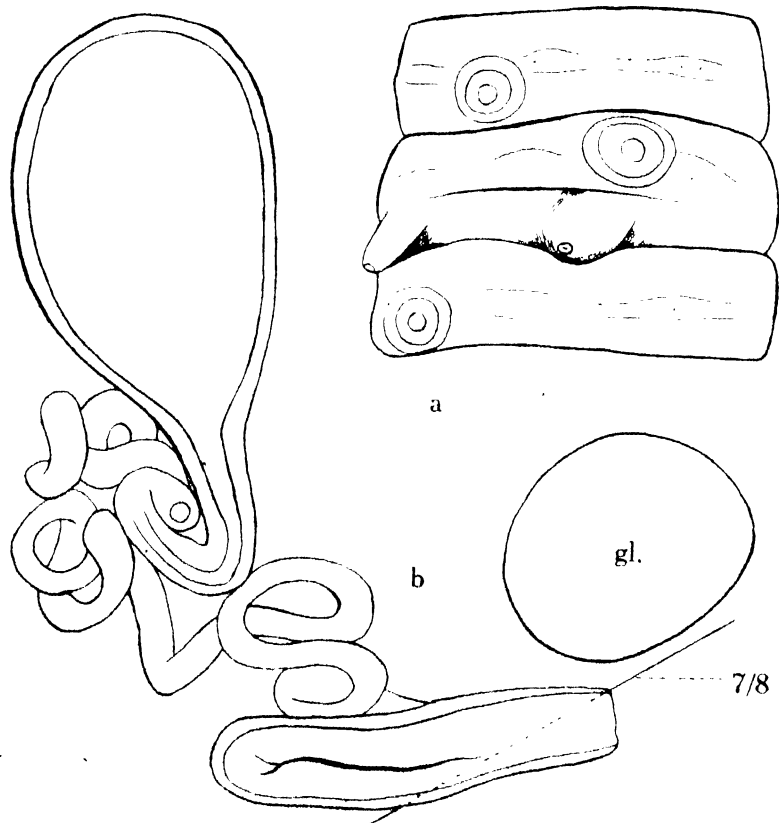


Fig. 1. *Drawida jeholensis*, n. sp. a, ventrolateral view of IX-XI, ca. $\times 18.9$. b, a spermatheca with atrium and an accessory gland, ca. $\times 70$.

edge of XII, in ab or b or just lateral to b, minute and not readily recognizable; the location of the pores can, however, be determined indirectly by the greyish translucent appearance of the epidermis surrounding the apertures.

Spermathecal pores are minute, forming longitudinal (in all the 10 specimens examined) slits on the posterior margin of VII, just medial to c; the region around the pore is slightly depressed in a crescent-shape.

Each genital papilla is spherical, forming a small and dully glistening tubercle surrounded by a whitish, smooth, thick and slightly elevated glandular rim of the body skin (Fig. 1, a). The papillae are found on VII-XI presetally or postsetally, between b and c or medial to b or midventrally; in most specimens, a single, small spermathecal papilla is found within each spermathecal depression, to be just lateral to the aperture. Their segmental position observed of 10 specimens at random is as follows:

Position	No. of specimens
on VII-XI & in 7/8	2
on VIII-XI & in 7/8	2
on VII-IX, XI & in 7/8	1
on VII-IX & in 7/8	1
on VII-X & in 7/8	1
on VII, IX, X & in 7/8	1
on VII, IX & in 7/8	1
on VIII XI	1

Internal anatomy:

Septa 5/6-8/9 moderately thickened, the rest thin; 10/11 is dorsally displaced posteriorly into the anterior part of XII and ventrally into about $\frac{1}{2}$ XI; the remaining ones are approximately normally inserted.

Gizzards are two or three in number, located in XII and XIII or in XI-XIII. Out of the 25 specimens opened, two in 15 specimens and three in 10 specimens. They are large and globular in shape; the first one is, in most cases, well-musculated even when three present, and slightly pushes anteriorwards the testis sacs. Last hearts in IX.

Testis sacs are one pair, of moderate size; each rather deeply constricted by 9/10, usually lying its smaller part in IX, 2-3.5 mm long and 1.3-2 mm broad. Sperm-duct is short with a few loose loops and passes into parietes just medial to prostate. Prostates are one pair, each rather small, thumb-shaped, usually slightly compressed, 1-2 mm long, narrowed ectally, slightly warty on surface. Central body is slenderly tubular and somewhat finger-shaped, about 0.6-1 mm long.

Ovarian chamber in XI is formed by 10/11 and 11/12; both septa are entirely fused dorsally but widely separated ventrally. All specimens are not yet completely mature; in both of ovarian chamber and ovisac, any free ova were not found (of 25 specimens opened). Ovisacs are slender and rod-like, and extend posteriorly into about XVI-XX. Possibly the ovisacs may become finger-shaped when they attained their maturity being filled with ripe ova.

Spermathecae with atria are found on the posterior face of 7/8 (Fig. 1, b). Ampulla is ovoidal or rounded, of about 0.7-1.2 mm diameter. Spermathecal duct is thin, moderate in length, of about 5-7 mm long, looped almost in its entire length, and not sharply marked off from the ampulla; it passes into the lateral side of atrium at about ental third, and its lumen is fused with the ectal end of the latter. Atrium is columnar, nearly equal in length to the diameter of ampulla.

Each accessory gland is tough-walled, ball-like in shape with a very short duct. The gland corresponding to the spermathecal papilla is usually found just in front of the septum 7/8 (Fig. 1, b).

Locality and material: Chihfen, 44 semi-clitellate and 8 aclitellate sps., September, '37, by Mrs. S. KOHASI and H. TADA.

Remarks:

In its external appearance the present species closely resembles *Dr. koreana*. In its internal features it closely resembles *Dr. koreana*, *Dr. japonica* and *Dr. propatula*. It is easily distinguishable from the last two mentioned by the large and rather conical male porophore and by its general body-shape; and from the first, which it most closely resembles, mainly by the distinctly longer and thinner spermathecal duct and also by the colouration of the body.

5. *Drawida gisti* MICHAELSEN 1931

1938 *Drawida gisti*, KOBAYASHI, Sci. Rep. Tôhoku Imp. Univ., Biol., XIII, 2, pp. 95-99, fig. 2.

Colour in formalin, uniformly yellowish grey with fleshy red clitellum. Length 120 mm, greatest diameter 5.8 mm, number of segments 205. Clitellum clearly differentiated from the neighbouring segments by its colour and thick glandulation, in IX $\frac{1}{2}$ -XIV; but, the colouration and glandulation on both segments IX and XIV are less distinct. Setae short, closely paired, those on II and clitellar segments delicate; setal distance $aa > bc$ anteriorly and $aa = bc$ posteriorly. Head of penis hardly

visible externally; posterior edge of the male pore (slit-like) slightly elevated as a lip. Spermathecal pore, in 7/8, in c-line. Genital papillae absent.

Three gizzards in XII-XIV. Testis sac constricted by 9/10, larger part lying in X, of about 2.8×2.3 mm. Prostate large, l-shaped, glandular portion about 8 mm, its duct r-shaped, slender, about 2.2 mm; penial pouch somewhat peach-shaped, about 2 mm; penis slender, blade-shaped, about 2 mm. Ovisac conical in shape, smooth on surface, fully filled with ripe ova, extending posteriorly into about XV. Spermathecal atrium very large, with characteristic urn-shaped gland which is a little larger than the ampulla.

Locality and material: Chihhsien, 1 mature sp.

Distribution: North China, Central Korea, South Manchoukuo.

6. *Drawida nemora* KOBAYASHI 1936

1938 *Drawida nemora*, KOBAYASHI, Sci. Rep. Tôhoku Imp. Univ., Biol., XIII, 2, pp. 99-102.

All the specimens at my hand are strongly contracted; body size is considerably variable. Hsinking-specimens (immature), about 65×5 mm in the largest one; in a complete Tumen-specimen, 124×9 mm and number of segments 247; a Fengtien-specimen which is, however, broken in the posterior part, is intermediate in size between the formers. Colouration is also variable: dorsally, yellowish blue in Hsinking-specimens, from dark to brownish blue with purplish and yellowish tinge in Tumen-specimens, dirty blue in Fengtien-specimen.

The following description is mainly based upon the mature specimens taken from Tumen. Setae on II and on the clitellar segments are smaller than those on the rest. Spermathecal pores are invisible externally, but the position is indirectly recognizable from the indistinct transversely crescent-shaped or slit-like depression which is found on the posterior edge of VII; in all cases, within the depression is found a light-coloured lip-like minute swelling. In all the present materials, gizzards are four in XII-XV. Yellowish brown, finely tubular masses are found on the dorsal vessel, accompanied by the blood sinuses. Testis sacs large, yellowish, about 5×3.6 mm; sperm-duct moderate in length, as thick as the spermathecal duct. Prostate poorly developed, with thin glandular investment, half buried in the parietal wall, discoidal in shape, about 2 mm in diameter; the terminal status of the male organ is similar to that of

the original specimens. Ovisac voluminous, containing a large amount of ripe ova, extending posteriorly into about XV. Spermathecal duct long, terminating without any trace of atrial dilatation just in front of 7/8; ampulla fairly large, ovoidal, about 4×3.5 mm; close to the ectal end of the duct is found usually a rather firmly-formed discoidal accessory gland.

Localities and materials: Hsinking, 10 immature sps., August, '34; Tonryan, near Fengtien, 1 immature sp., August, '37, Mr. K. KOBA; Tumen, 3 mature sps., August, '38.

Distribution: Central and North Korea, Central Manchoukuo.

Family Megascolecidae

Genus *PHERETIMA* KINBERG em. MICHAELSEN 1900

7. *Pheretima hupeiensis* (MICHAELSEN) 1895

1938 *Pheretima hupeiensis*, KOBAYASHI, Sci. Rep. Tôhoku Imp. Univ., Biol., XIII, 2, pp. 152-153.

Most of both Fengtien- and Tashihchiao-specimens are heavily infected by Gregarine parasites, but both Harbin- and Chihhsien-specimens are all apparently healthy. Testis sacs of X and XI are either annular or vertically U-shaped.

Localities and materials: Harbin, 5 clitellate sps. and several fragments; Fengtien, 9 a clitellate and juvenile sps.; Tashihchiao, 6 clitellate and 1 a clitellate sps.; (Dairen, based upon a verbal communication from Mr. J. ÔIZUMI); Antung, several clitellate and a clitellate sps., August, '35.

8. *Pheretima aggera* KOBAYASHI 1934

(Fig. 2)

1938 *Pheretima aggera*, KOBAYASHI, Sci. Rep. Tôhoku Imp. Univ., Biol., XIII, p. pp. 153-155, fig. 13, a.

Description:

External characteristics:

Body long with numerous segments; generally longer than the Korean specimens. Length 175-298 mm, greatest diameter 5.5-10 mm, number of segments 150-171. Colour in formalin: dorsally dark brown and concentrated middorsally and preclitellarly, ventrally light brown or sometimes rather pale, clitellum chocolate. First dorsal pore in 12/13, distinct and functional; an indistinct and non-functional pore-like marking may be

rarely found in 11/12. Setae small, slightly enlarged on both preclitellar and posterior segments; ventral ones on preclitellar segments may be irregularly interrupted. No marked difference in size and also in interval is found between ventral and dorsal ones. Both mid-dorsal and -ventral breaks present but slight, approximately $aa=1.3-2$ ab and $zz=1.5-2.5$ zy . Setal number subequal to that of the Korean specimens; spermathecal setae 20-23/VIII, male pore setae 16-21. Of the male pore setae, 3-4 are always found on the medial part of male disc, and of those, the extreme lateral one is usually smaller than the rest and is often not clearly visible externally.

Crescent-shaped secondary male pores with moderate elevation are found on ventrolateral surface of XVIII; the body wall lateral to the pore is light-coloured, prominently elevated and lacks the setae. Within the shallow copulatory chamber is found a wrinkled and glandulated male disc, on which 3-4 setae are planted. On the disc, at the extreme lateral side which is slightly lower than the medial part or often slightly sunken into the coelom, is found a very small, transversely club-shaped male porophore (Fig. 2, a). The porophore is anteriorly and posteriorly provided with two very small oval brownish yellow papilla-like swellings which are half buried into the ground tissue. Male pore opens as a minute aperture at the lateral part of the club-shaped porophore. These porophore and papilla-like swellings form a small but firm, laterally directed arrow-head-like body; this body may be more or less variable in form corresponding to the degree of the contraction of the specimens (Fig. 2, b-d). This structure is definite in existence, though it has not been described in my previous papers. In all the Manchoukuo specimens, no genital papillae are found on the large raised part of the disc medial to the porophore. Frequency of the occurrence of these papillae was examined using the Korean specimens. They are 0-4 in number on each disc; it is not rare that they are totally absent on both sides, and on the contrary, it is rather rare that they are 4 on each disc. When a single papilla exists, it resembles somewhat that of *Ph. tschiliensis*, but is much smaller. *Ph. aggera* which was described in my latest paper ('38) in reality contains two species of the present species and *Ph. tschiliensis*; the Sainei-specimens on which I made a special remark and gave an illustration in that paper, concern to the latter species.

Spermathecal pores, three pairs in 6/7-8/9; each situated on a small papilla-like swelling which is found within a large, eye-shaped depression. Both anterior and posterior borders of the depression is moderately swollen.

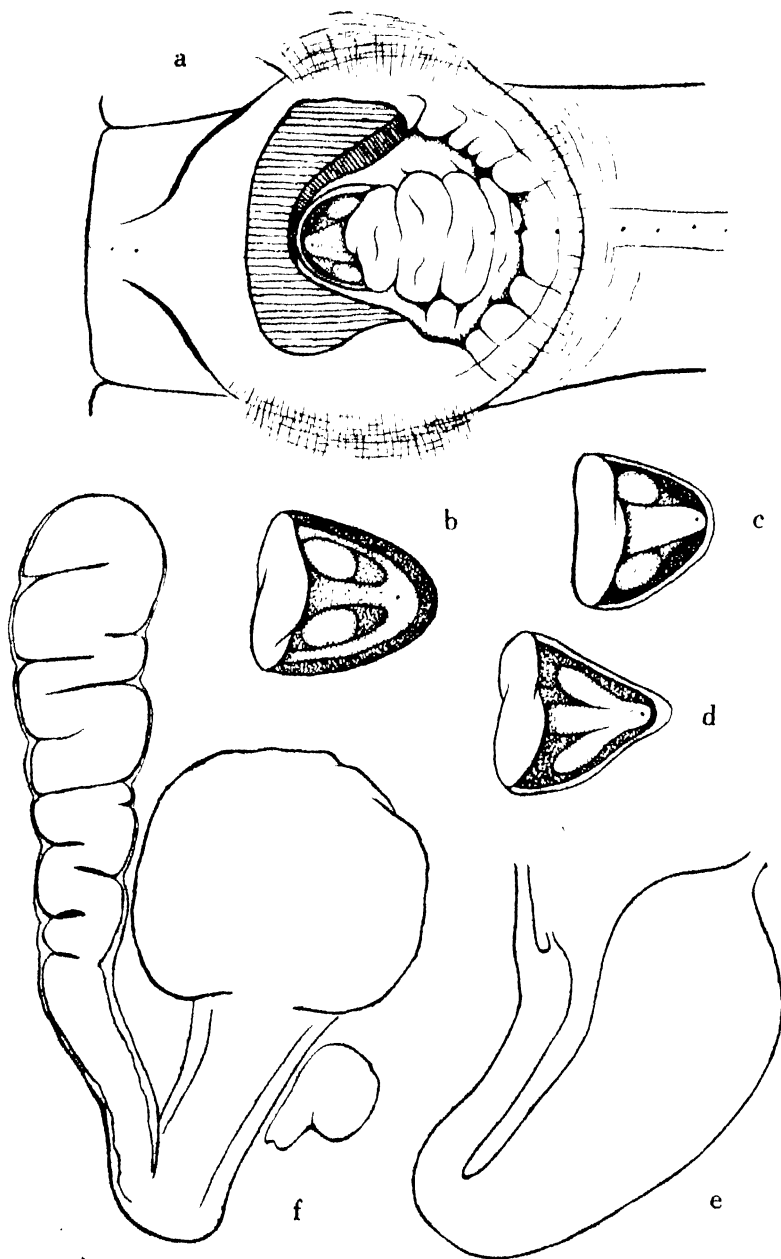


Fig. 2. *Pheretima aggera* KOBAYASHI. a, ventrolateral view of male pore region of one side (cut off the lateral wall to show the surface of the male disc); e, a prostatic duct; f, a spermatheca with an accessory gland of the genital papilla; a, e & f, ca. $\times 18.9$. b-d, variation of the arrow-head-like male pore, here with two papilla-like swellings, drawn with free hand.

When the depression is closed, the pore is invisible externally. To the spermathecal depression anteriorly or posteriorly or both, one or two very small simple swelling-like genital papillae are often found on VI-IX or on some of them. Size, and frequency of occurrence of the papillae are similar to those seen in the Korean specimens. As the papillae of the present species are clearly different in shape, size and also in position from those of *Ph. tschiliensis*, these two species may be easily distinguishable from each other only by these features of this structure.

Internal anatomy:

Septum 8/9 ventrally present but thin, 9/10 absent. Intestine begins to swell in XV. Caeca simple, in XXVII, extending anteriorly into about XXII, large and long, finger-shaped with broad basal portion, each ventrally with several serriformed outgrowths. Hearts in X-XIII, fairly large; those in X subequal in calibre to those in XI-XIII. Lymph glands small, club-shaped, found behind caecal segment caudalwards.

Seminal vesicles moderate in size or often rather small, circular or oval in shape, vesicular on surface, each with a moderately-sized, distinctly constricted, ovoidal, smooth dorsal lobe; usually the anterior pair of vesicles are smaller than the posterior. Testis sacs ventral in position and moderate in size; the anterior pair forming a low or a very low U-shaped sac and the posterior pair a quadrate sac. Pseudovesicles, one pair, moderate in size, club-shaped, behind 12/13. Prostate gland small, occupying only two or more of segments, in XVIII- or XVII $\frac{1}{2}$ -XIX, divided into many finger-shaped pieces. Duct usually curved in a U-like manner, the ental half is slender being of nearly equal thickness while the ectal half is very thick and muscled, being about 5 or more times as thick as the former (Fig. 2, e). With the narrowed ectal end, the duct enters, through a cushion-like, moderately-sized glandular tissue of the male disc, into the arrow-head-like body, and opens at the tip of the porophore within the shallow copulatory chamber; on both sides of the ectalmost of the duct, two, very small, oval shaped, apparently soft accessory glands are recognizable embedded in the tissue without stalk.

Spermathecae large (Fig. 2, f). Ampulla rounded, sometimes uneven or irregularly wrinkled on surface. Duct thick and muscled, slightly narrowed in the ectal part, nearly equal in length to the ampulla, sharply marked off from the latter. Diverticulum arising from the ectal end of the ampullar duct, always longer than the main portion; the ectal portion which serves as duct, is short and subequal in length to the ampullar duct which is thick-walled and nearly straight, while the remaining longer

ental portion is thin-walled, slightly dilated, coiled into several compact loops running in zigzag manner and is inclosed within a delicate sheath. Small stalked accessory glands are found inside corresponding to genital papillae externally present.

Localities and materials: Chihhsien, 8 clitellate and 7 a clitellate sps.; Tashihchiao, 15 clitellate and 11 a clitellate and juvenile sps.; Dairen, 9 clitellate and 3 a clitellate sps., May, '37, by Mr. J. ÔIZUMI; (Chihfen, 5 a clitellate and juvenile sps., Sept., '37, by Mrs. S. KOHASI and H. TADA); (Sanjuriho, Chinchow, Pitzewo, Kakuchingpo, Taiseishan, in Kwanto Province, based upon a verbal communication from Mr. J. ÔIZUMI).

Distribution: Central and South Korea, South Manchoukuo.

Remarks:

It is noteworthy that the genital papillae found on the male disc are absent in all cases of the Manchoukuo specimens, though they are often present in the Korean specimens.

The present species is specifically distinct from *Ph. tschiliensis*, though in many respects they are closely allied. The main differences existing between them may be found in the following points: (1) the minute structure of the terminalia of the male genitalia, (2) the size, shape and relative position of genital papillae found in the spermathecal region, and (3) the size and shape of both seminal vesicle and prostate (with duct). *Ph. asiatica*, *Ph. tibetana*, *Ph. yamadai*-A-form (CHEN '33), *Ph. grahami*, *Ph. praepinguis*, *Ph. tschiliensis* and the present species very closely resemble one another in many respects. But, each of the first five species appear to be more closely allied to *Ph. tschiliensis* than to the present species. But, as we have not any really accurate information about these five species, a re-examination of them is very desirable, and at present, I am therefore not able to give here any further particulars concerning the synonymy or dissimilarity between these species.

All the present specimens are apparently free from parasites. In the younger specimens from Chihfen, the spermathecal pores, genital papillae and prostatic duct are very poorly developed, though they are characteristic in shape and in position. Judging from these features, they may be probably identical with the present species.

9. *Pheretima tschiliensis* MICHAELSEN 1928

(Fig 3)

- 1930 *Pheretima kiangsuensis*, CHEN, Sci. Rep. Centr. Univ. Nanking, I, 1, pp. 24-28, figs. 7-9.
- 1931 *Pheretima tschiliensis*, MICHAELSEN, Lingnan Sci. Jour. VIII, p. 160.
- 1931 *Pheretima tschiliensis*, MICHAELSEN, Peking Nat. Hist. Bull., V, 3, p. 2.
- 1931 *Pheretima kiangsuensis*, CHEN, Contr. Biol. Lab. Sci. Soc. China, VII, 3, pp. 119-122, fig. 1.
- 1933 *Pheretima tschiliensis*, CHEN, *ibid.*, IX, 6, p. 250.
- 1935 *Pheretima tschiliensis*, GATES, *Smithon.*, Misc. Coll., XCHI, 3, pp. 16-18, figs. 13 & 14.
- 1938 *Pheretima tschiliensis*?, KOBAYASHI, Sci. Rep. Tôhoku Imp. Univ., Biol., XIII, 2, p. 154, fig. 13, b.
- 1939 *Pheretima tschiliensis*, GATES, *Proc. U. S. Nat. Mus.*, 85, pp. 488-494*.

Description:

External characteristics:

Length 154-197 mm, greatest diameter 7-8 mm, number of segments 140-145. Colour, dorsally brownish blue anteriorly, purplish brown posteriorly, ventrally dusty grey or pale, clitellum dusty chocolate; in the specimens preserved in formalin, the bluish colour is faded away. Prostomium, epilobous ca. 1/2; moderate in length and in width. First dorsal pore in 12/13, distinct and functional; frequently, it may be non-functional though is distinct. Clitellum entire, in XIV-XVI, without setae.

Setae moderate in size, beginning on II; usually those on II-IX slightly enlarged; no marked difference in size and also in interval is found of the ventral and dorsal ones. Middorsal breaks very slight if present, and midventral ones slight, approximately $aa=1.5-2.5\ ab$. Setal number taken from several specimens is as follows: 38-43/III, 50-54/V, 54-56/VI, 53-57/VIII, 57-60/IX, 59-61/XII, 57-58/XIII, 63-69/XXV, spermathecal setae 20-22/VI, 21-24/VII, 22-26/VIII, 23-28/IX, male pore setae (5-7) + (10-13) + (5-6) = 21-24 (5-7 on disc).

When the copulatory chamber is retracted, the general appearance of XVIII is very close to that of *Ph. aggera*; but, when a large genital papilla is exposed in various degrees from the mouth of the chamber, as its appearance is characteristic we may easily identify the present species. Within the shallow copulatory chamber provided with a crescent-shaped secondary pore, is found a wrinkled raised male disc, on which 5-7 setae are planted and also a single presetal (constantly) fairly large circular and centrally depressed genital papilla is always placed (sometimes, this papilla is rather raised from the surface of the disc). On the disc, at the extreme lateral side which is slightly lower than the medial larger part, is found a very small circular male porophore. Anteriorly and

* I received this paper after the completion of this manuscript.

posteriorly to the male porophore are found two (rarely, one on one side), very small but distinctly demarcated, circular and centrally depressed genital papillae. These porophore and genital papillae are, as a whole, in the form of a small but firm body which may be protrusible (Fig. 3, a).

Spermathecal pores, three pairs in 6/7-8/9; each representing in a very minute aperture in the intersegmental furrow, just anteriorly with a shallow crescent-shaped depression containing one or two indistinctly demarcated lip-like swellings and just posteriorly with a light-coloured crescent-shaped ridge. When the region is sunken into the intersegmental

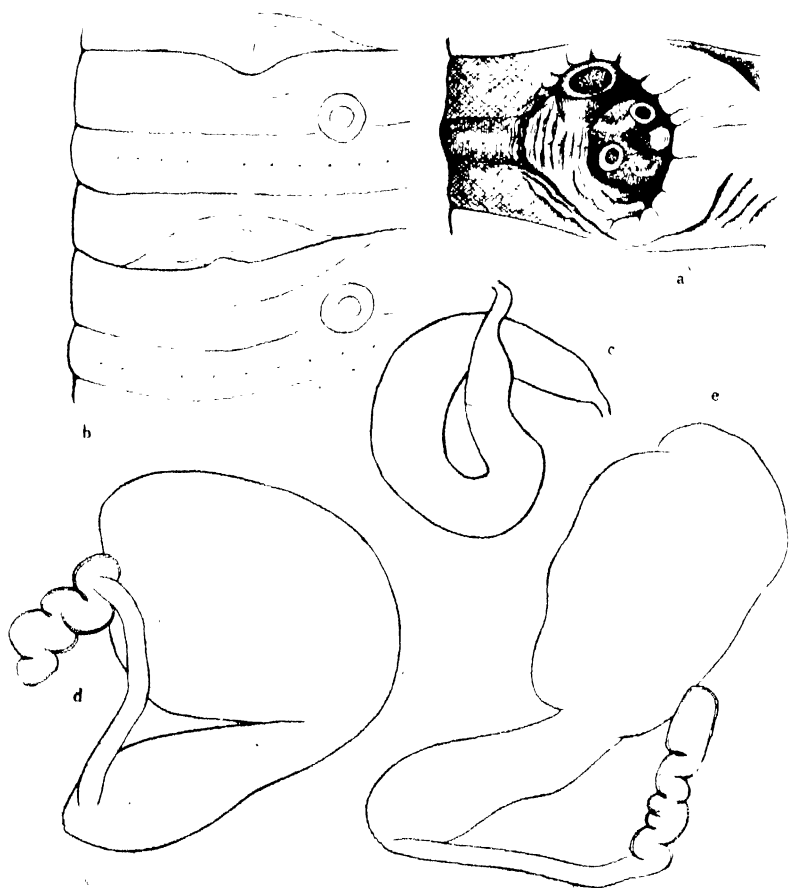


Fig. 3. *Pheretima tschiliensis* MICHAELSEN. a, ventrolateral view of male pore region of one side (copulatory chamber is half everted and a large genital papilla on male disc and male porophore with two very small genital papillae are seen externally); b, spermathecal region (VII & VIII); c, a prostatic duct; d & e, spermathecae; a-e, ca. $\times 12.6$.

furrow, both the aperture and the depression become invisible externally; even in such case the borders anterior and posterior to the pore are swollen. In all cases no spermathecal papillae are found. Genital papillae, three pairs on VII-IX, always presetal along the setal zone; each medial to the spermathecal pore-line with about 3-5 setal distance, circular and centrally depressed, slightly smaller and less distinct in central depression than the larger papilla on the male disc. They occur rather constantly, though some of them are sometimes absent; of these three pairs, those on VIII are most constantly found (Fig. 3, b).

Internal anatomy:

Septa generally well-developed; 5/6-7/8, 10/11 and 11/12 much thickened, 12/13 less thickened, 8/9 ventrally thin and is found between the last pair of the spermathecae, 9/10 absent. Gizzard behind 7/8, globular in shape; intestine begins to swell in XV. Caeca simple, large and long, horn-shaped, in XXVII, extending anteriorly into about XXII or XXIII, each ventrally with several serriformed outgrowths. Hearts in X-XIII; those in X are large, but sometimes they are bound to the anterior face of 10/11. Lymph glands large or moderate in size, lobular, found behind caecal segment caudalwards.

Seminal vesicles, vesicular, voluminous, fairly large, occupying all the interior of the respective segment and dorsally meeting with one another; each with a large dorsal lobe. Dorsal lobe of the anterior pair may sometimes be subdivided into two or three smaller ones. Testis sacs ventral in position, massive and large; the anterior pair forming a U-shape and the posterior a transverse sac. Pseudovesicles, one pair, small or sometimes very small, behind 12/13.

Prostate gland large, usually in XVI-XXI, divided into many finger-shaped pieces. Duct long, looped in a hair-pin-shape; entally rather thin and ectalwards gradually increasing the thickness and muscularity (Fig. 3, c). Ectal end of the duct becomes thinner and enters, through a fairly large cushion-like glandular tissue, into a firmly-formed body situated at the bottom of the copulatory chamber, and opened at the tip of the porophore. Accessory glands with solid stalks are found close to the ectal end of the prostatic duct corresponding externally to the smaller papillae. A similar gland but of larger size, corresponding to the larger papilla, is found near these.

Spermathecae fairly large, three pairs in VII-IX. Ampulla large, ovoidal, with smooth surface. Duct thick, slightly shorter than the diameter of ampulla, not sharply marked off from the latter, slightly

enlarged at the ectal part. Diverticulum subequal to, or a little shorter than, the main portion; the ectal half slender but thick-walled, entering into the ampullar duct at its ectal end close to the parietes, and the ental half thin-walled, slightly dilated, coiled into several compact loops running in a zigzag manner; a delicate sheath present around these but is rather indistinct (Fig. 3, d & e). No accessory glands are found close to the ectal end of the ampullar duct. Slightly posteriorly to each spermatheca is found a large accessory gland with stalk; its glandular portion is often divided into two or more numbers of smaller lobes.

Locality and material: Chihhsien, 15 clitellate and 22 aclitellate and juvenile specimens.

Distribution: North and Central China, South Manchoukuo, Central Korea.

Remarks:

Ph. tschiliensis is placed, at present, systematically in a confusing manner. In the original paper, it was inadequately described by MICHAELSEN ('28). According to GATES ('35), CHEN's *Ph. yamadai*-A-form ('33), MICHAELSEN's *Ph. asiatica* ('00) and *Ph. tibetana* ('31) are all synonymous with the present species; if this is really the case, the present species and the others above mentioned are synonymous with *Ph. asiatica*. According to CHEN ('36), GATES' *Ph. grahami* and *Ph. praepinguis* ('35) are also synonymous with the present species. Re-examination of these questionable and little known species is very desirable.

Ph. tschiliensis (= *Ph. kiangsuensis*) has been most adequately characterized by CHEN ('30 & '31). His Nanking- and Soochow-specimens are different in some characteristics from the Omei-ones (it is questionable to me whether they are specifically identical or not). According to his description, the occurrence, number and size of spermathecal papillae (not genital papillae: see, '38, KOBAYASHI) are variable in the Chinese specimens; his descriptions are as follows: in the Nanking- and Soochow-specimens ('30) there is "anteriorly an ampulla-like round topped papillae which, in some cases, is absent.... Also the small glands around spermathecal region may be absent", and in the Omei-specimens ('31) there is "Skin around spermathecal pores usually strongly swollen and glandular in appearance; pale in colour, often wrinkled around the pore;... Two or more large button-shaped papillae situated posterior and anterior to each pore (largest one about 1.5 mm in diameter)", and in the re-examination ('33) of the MICHAELSEN's type "spermathecal pores with genital papillae". In the present study, with regard to the presence or absence

of the spermathecal papillae a careful examination was made (and likewise of the accessory gland), but none of them was found in any of the Manchoukuo specimens.

I think that the constant occurrence of a large presetal genital papillae on the male disc has a systematic value. In fact, except CHEN's Kwanhsien specimen, its occurrence is constant in the Chinese specimens. One smaller, postsetal genital papilla on the male disc of some of the Nanking- and Soochow-specimens described by CHEN, is absent in all the Manchoukuo specimens. Considering the description and figures given by CHEN, his Nanking- and Soochow-specimens of *Ph. tschiliensis* seems to contain two species of the present species and *Ph. aggera*.

Judging from CHEN's description ('30), the relatively smaller size of the seminale vesicles, of the prostate glands and of the spermathecae in his Nanking- and Soochow-specimens may be caused on the one hand, at least in some degree, by infection from parasites, or on the other hand by the fact that his specimens partly included *Ph. aggera* which have those organs smaller. The present specimens are apparently free from parasites, and as in his Omei-specimens, and in some of the Nanking- and Soochow-ones, the seminal vesicles and spermathecae are fairly large and also the prostate glands.

Occurrence of the present species in Central Korea is shown in the paragraph relating to *Ph. aggera* in the present paper.

Family Lumbricidae

Genus *EISENIA* MALM 1877 em. MICHAELSEN 1900

10. *Eisenia nordenskiöldi* (EISEN) forma *typica* 1879

1910 *Helodrilus (Eisenia) nordenskiöldi typica*, MICHAELSEN, Ann. Mus. Acad. Imp. Sci. St.-Petersburg, XV, pp. 17-18.

1924 *Eisenia nordenskiöldi typica*, SVETLOV, Bull. Inst. Rech. Biol. Perm, 2, p. 322.

1929 *Eisenia nordenskiöldi typica*, MICHAELSEN, Ann. Mus. Zool. Acad. Sci. URSS., XXX, p. 329.

Description:

Length 50-110 mm, greatest diameter 3-6 mm, number of segments 124-165; generally in the range of 130-145. In spite of a large number of worms were obtained, the complete ones were rather small in number. Colouration somewhat resembles that of *E. foetida*. In fully-extended worms, intersegmental furrows may appear yellowish, but they do not show any banded appearance as in the case of the latter species. Dorsally

deep or slightly darkened violet-red, ventrally whitish to yellowish grey; dorsolateral surface of IX-XI faded; clitellum fleshy or sometimes yellowish flesh.

Prostomium, epilobous ca. $1/3-1/2$. First dorsal pore in $4/5$, distinct and functional.

Setae moderate in size, closely paired, $ab > cd$ and $aa > bc$ throughout the entire length of the body, or $aa = ca$, 7-8 $ab = 1\frac{1}{2}-1\frac{2}{5} bc$, dd a little or a very little smaller than $1/2$ of the circumference. Setae ab of about XXV XXXV or most of them and cd of X-XII or -XIII may be planted on whitish indistinct tumescences. These genital setae are not large being about 0.5-0.6 mm long, proximally curved slightly, distally nearly straight but provided with a keel-like curvature and with longitudinal grooves in its about distal $1/4-1/5$.

Clitellum saddle-shaped, usually in XXVI or XXVII-XXXIII ($=7$ or 8); seldom $1/_{n}XXVI-XXXIII$ or $XXVI-1/_{n}XXXIII$, or very seldom XXVI-XXXII or XXVII-XXXII ($=6-7 1/_{n}$). In all the Chiamussu-specimens, it extends from XXVII-XXXIII. In the others, such constancy is not found. Pubertatis tubercle, usually in XXIX-XXXI, seldom in $\frac{1}{3}XXXVIII-XXXI$ or XXIX- $\frac{2}{3}XXXI$ or $\frac{1}{3}XXXVIII-\frac{2}{3}XXXI$; it is usually of a distinct groove immediately lateral to b-line, sometimes it may be of a slightly elevated 'ridge', or seldom, it may be rather indistinct.

Male pores on XV, usually without marked glandular elevation, between b and c , nearer to b .

Spermathecal pores, two pairs in $9/10$ and $10/11$, close to middorsal line.

No septa specially thickened; $6/7-8/9$ slightly thickened. Crop in XV-XVI; gizzard in XVII-XVIII.

Testes and funnels free, in X and XI. Seminal vesicles, four pairs in IX-XII; the anterior two pairs on the anterior faces of $9/10$ and $10/11$, and the posterior two pairs on the posterior faces of $10/11$ and $11/12$. Vesicles in IX and X moderate in size and usually simple in feature; those in X are slightly (or sometimes clearly) smaller than those in IX. Those in XI and XII are larger than the anterior two pairs, and are in most cases slightly lobated.

Spermathecae small and ball-like, each provided with an inconspicuous stalk.

Localities: Hailar, Ilikete, Tsitsihar, Keshan, Harbin, Chiamussu, Iyasaka, Poli, Mutanchiang, Tumen, Kirin, Hsinking, Fengtien, Fsfeng, Chihnsien.

Distribution: Siberia, E.S.-Russia, Manchoukuo. (N.-Europe, N.-America, Hawaiian Island doubtful).

Remarks:

The present species is the commonest earthworm found in North Manchoukuo. Its general appearance resembles that of *E. foetida*. It is noteworthy that in all the Chiamussu-specimens the clitellum extends from XXVII-XXXIII, though in the others no such constancy was found, and that its extension agrees with that of the specimens which have been reported from Siberia and Eastern Russia. With regard to the shape of the pubertatis tubercle, the two types were found to be nearly the same as that given in MICHAELSEN's work ('10 a); but the usual type of the Manchoukuo specimens agrees with that of N.E.-Siberian ones. In most cases in the soil, in which the worms lived, a number of cocoons were found at the depth of about 10-20 cm.

11. *Eisenia nordenskiöldi* forma *manshurica*, n. subsp.

Description:

Colour in formalin: dorsally, from dusty red to blackish red with a faint purplish tinge, preclitellarly concentrated, and lateral and dorsolateral surface of IX-XI faded; ventrally dusty grey; clitellum deep purplish or dusty brown; its colouration is clearly different from that of f. *typica*. Length 111-144 mm, greatest diameter up to 6.5 mm, number of segments 154-175. Prostomium, epilobous ca. 1/3. First dorsal pore in 4/5.

Setae are generally a little larger than those of f. *typica*; setal distance $ab > cd$, $aa > bc$, dd a little shorter than $1/2$ of the circumference. Setae ab of XXII-XXXV (or most of them) slightly enlarged and planted on very small whitish tumescences; cd of XIV-XXXIV (or most of them) and ab of V-IX may be also slightly enlarged, but are not so markedly enlarged and are not planted on the tumescences as in the case of the former setae. Genital setae are about 0.8 mm long, proximally only slightly curved; the distal about $1/4$ slightly thinner than the proximal and is provided with longitudinal grooves (examined a ventral seta from the clitellar region).

Clitellum saddle-shaped, in XXVI- or XXVII-XXXIV (=8-9). Pubertatis tubercle in XXIX-XXXII; it is of a distinct groove, and is placed immediately lateral to b-line.

Male pores on XV, without marked glandular elevation, between b and c, nearer to b; its general appearance closely resembles that of f. *typica*.

Seminal vesicles, four pairs; the anterior two are small and simple in shape; those in X are a little smaller than those in IX. The posterior two are much larger than the anterior ones, and each of the former is divided into 2-3 lobules in the anterior face.

Spermathecae ball-like, each with an inconspicuous stalked portion which may be slightly longer than that of f. *typica*.

Localities and materials: Chihhsien, 4 semi-clitellate sps.; Anshan, 2 clitellate and 2 a clitellate sps.; August, '38, by Mr. KUNIO SUDA; Mutanchiang, 2 semi-clitellate sps.

Remarks:

The present form stands related more closely to f. *lagodechiensis* than to f. *typica*, in the longer extension of the clitellum and in the features of the pubertatis tubercles. But, it differs from f. *lagodechiensis* in being destitute of the glandular elevation around the male pore, in the colouration of the body and in the relative size of the seminal vesicles. The male pore is similar to that of f. *typica* in general aspects.

12. *Eisenia rosea* (SAV.) forma *typica* 1826

1924 *Eisenia rosea*, SVETLOV, Bull. Inst. Rech. Biol. Perm, II, p. 322.

1935 *Eisenia rosea typica*, CERNOSVITOV, Monogr. tschechosl. Lumb., p. 37.

1936 *Eisenia rosea*, KOBAYASHI, Sci. Rep. Tôhoku Imp. Univ., Biol., XI, 1, p. 183.

1937 *Eisenia rosea*, ČERNOSVITOV, Rec. Ind. Mus., XXXIX, 2, 107.

1937 *Eisenia rosea typica*, TÉTRY, Bull. Mus. Hist. Nat., IX, p. 143.

1938 *Eisema rosea*, TÉTRY, *ibid.*, X, p. 74.

Localities and materials: Keshan, 3 clitellate sps.; Fengtien, 1 clitellate sp., June 27, '37, by Mr. K. KOBAYASHI.

Distribution: Whole Europe, Syria, Transcaucasia, Turkey, Siberia, N. & S. India, Egypt, Morocco, Canary Islands, Azores, Kapland, Catham Island, N. & S. America, New Zealand, Japan, Korea, Manchoukuo.

13. *Eisenia rosea* (SAV.) forma *macedonica* (ROSA) 1893

1926 *Allolobophora (Eisenia) rosea macedonica*, PICKFORD, Nat. Hist. Wicken Fen, III, p. 227, fig. 2, B & D.

Localities and materials: Keshan, 3 clitellate sps.; Nanhkingan, slightly distant from the political boundary of Inner Mongolia, 1 clitellate sp.; Tumen, 1 clitellate sp.; Mutanchiang, 1 clitellate sp.; Chaoyangchen, 2 clitellate sps.

Distribution: Macedonia, Switzerland, England, Manchoukuo.

A note on *E. rosea* f. *typica* and f. *macedonica*.

Eisenia rosea typica is a form of the world-wide distribution. *E. rosea macedonica* has been found only in Macedonia, Switzerland and England. PICKFORD ('26) reported these two forms and their intermediate form from Wicken Fen. F. *macedonica*, MICHAELSEN described in his Tierreich "Gürtel von 25. order 26.-31, 32. order 33. Seg. (=6 bis 8). Ventral Borstenpaare des 26.-33. Segm. auf kleinen papillen am Rande des Gürtels. Im übrigen gleich der typischer Form.", and PICKFORD described in her paper "In f. *macedonica*, which has ventral papillae in the region of the clitellum, there were usually both lateral and ventral papillae irregularly disposed between segments 8-13. In f. *typica* these were normally absent, and ventral papillae frequently occurred on or about segment 24. . . . A number of specimens of intermediate character occurred, the number of clitellar segments bearing copulatory papillae and the size of these papillae varying greatly." Thus, there are found some slight differences concerning the genital papillae between the English and the continental specimens; the number and position of genital papillae which have been treated as the criteria for these two forms of this species vary considerably.

In the Manchoukuo specimens, the number and position of the genital papillae also vary considerably, and there are found two forms and an intermediate form as with PICKFORD's specimens. It seems to me, judging from the present examination, that it is not necessary to separate these two forms. However, when considering the characteristic status of their distribution, the separation of the forms above mentioned appear to have some significance from the taxonomic viewpoint. Thus, a further study of these two forms is much to be desired.

The number and position of the genital papillae examined in the two forms and in the intermediate forms are indicated below.

According to PICKFORD, it is said that in Wicken Fen, f. *macedonica* is much less numerous than f. *typica*. It is noteworthy that most of the Manchoukuo specimens belong to f. *macedonica* though the total number examined is very small. If the separation of these two forms is essential, the occurrence of f. *macedonica* in Manchoukuo has an important significance in concerning the distribution of the family Lumbricidae. In the tendency of the superpapillation, f. *macedonica* of Manchoukuo appears to be more closely related to the forms found in Macedonia and in Switzerland than to those in England.

No.	Locality	I. f. <i>typica</i>		No. of specimens
		Right	Left	
1	Keshan	0	0	1
2	..	cd of 9 & 10	cd of 10 12	1
3	..	cd of 9-12	cd of 9 & 10	1
II. f. <i>macedonica</i>				
1	Keshan	cd of 9 ab of 24-33	0 ab of 24-33	1
2	..	cd of 10-12 ab of 27-34	cd of 10-12 ab of 27-34	1
3	..	cd of 10 12 ab of 27-34	0 ab of 27 34	1
4	Nanhkingan	cd of 9 ab of 25-33	cd of 9 ab of 25 33	1
5	Tumen	cd of 10 ab of 29-31	cd of 11 ab of 29-31	1
6	Chaoyangchen	ab of 25, & 30-32	ab of 31 & 32	1
7	..	ab of 27-32	ab of 27-32	1
8	Mutanchiang	cd of 10-12 ab of 19-13 ab of 24 32	cd of 11 & 12 ab of 11 13 ab of 24-32	1
III. Intermediate form				
1	Keshan	ab of 8 cd of 10 & 11 ab of 27	ab of 8 cd of 10 & 11 0	1

14. *Eisenia foetida* (SAVIGNY) 1826

1924 *Allolobophora foetida*, SASAKI, Sci. Rep. Tôhoku Imp. Univ., Biol., I, 1, pp. 89-90.

1935 *Eisenia foetida*, ČERNOSVITOV, Monogr. tschechosl. Lumb., pp. 34-36. (Literature: see this paper).

1935 *Eisenia foetida*, KOBAYASHI, Zool. Magaz., 47, p. 130.

1936 *Eisenia foetida*, KABURAKI and MISAKA, Zool. & Botany in Nikko, p. 514.

1936 *Eisenia foetida*, NOMURA and KOBAYASHI, Zool. Magaz., 48, pp. 885-893.

1937 *Eisenia foetida*, ČERNOSVITOV, Rec. Ind. Mus., XXXIX, 2, p. 107.

1937 *Eisenia foetida*, TÉTRY, Bull. Mus. Hist. Nat., IX, p. 143.

1938 *Eisenia foetida*, TÉTRY, ibid., X, p. 74.

1938 *Eisenia foetida*, KOBAYASHI, Ann. Zool. Japon., XVII, 3-4, p. 415.

1938 *Eisenia foetida*, KOBAYASHI, Jour. Chosen Nat. Hist. Soc., XXIV, pp. 6-18.

Locality and material: Dairen, 1 clitellate and 1 a clitellate sps., by Mr. J. ÔIZUMI.

Distribution: Whole Europe, Japan, Korea, S. Manchoukuo, N. & S. America, Canary Islands, Azores, Madeira, Bermuda Islands, Africa, Hawaii, New Zealand, Australia, India, Burma.

Genus ALLOLOBOPHORA EISEN 1874 em. ROSA 1893

15. *Allolobophora hataii*, n. sp.

(Figs. 4 & 5)

Description:

Length 78-97 mm, greatest diameter 2-3.2 mm (in clitellar region), number of segments 134-142. Except the clitellum, segments behind male pore region with 3 annuli. Colour uniformly grey except the clitellum which is light yellow or light brownish yellow.

Prostomium proepilobous (Fig. 4, a). First dorsal pore in 4/5, sometimes in 5/6.

Setae closely paired; setal distance aa is nearly twice as bc; ab is greater than cd; dd is smaller than $1/2$ of the circumference; aa:ab:bc:cd:dd: $1/2 \mu = 96:7:46:5:175:194$ observed on a segment immediately posterior to the clitellum. Setae cd of X, XI and XII, and ab of XV, XVI and XXV-XXXII are situated on papillae; ab of IX may rarely be also situated on papillae. Of these papillae, those on XV and XVI are prominent and are constantly found (Fig. 4, b & c; Fig. 5).

Clitellum saddle-shaped, in XXIV-XXXII; it is not rare that it invades anteriorly XXIII and posteriorly XXXIII (=9 or a little more than 9). Pubertatis tubercle in XXIX-XXXI, ridged and distinct in outline, immediately lateral to the papillae in position (Fig. 5).

Male pores are seated on very prominent and somewhat irregularly circular papillae, on XV, encroaching anteriorly about half on XIV and posteriorly about one third on XVI. On each side of XV, a papilla with setae ab is coalesced with the male pore-papilla, and setae cd are situated on the dorsal part of the latter (Fig. 4, b & c; Fig. 5).

Female pores were not definitely identified.

Spermathecal pores are absent.

Septa 6/7, 7/8 and 8/9 moderately, 9/10 slightly thickened, the remaining ones thin. Crop in XV-XVI; gizzard in XVII-XVIII.

Testes and funnels are free in X and XI. Seminal vesicles four pairs in IX-XII, darkened in colour; the posterior two pairs are larger than the anterior ones, and are either nodular in appearance or consist of a few small spherical lobules; those in X are nearly equal in size to those in IX.

Spermathecae are entirely absent.

Setae cd of X-XII, and ab of XV, XVI and XXV-XXXII (rarely, ab of IX also), are found in the internal dissection to be contained in

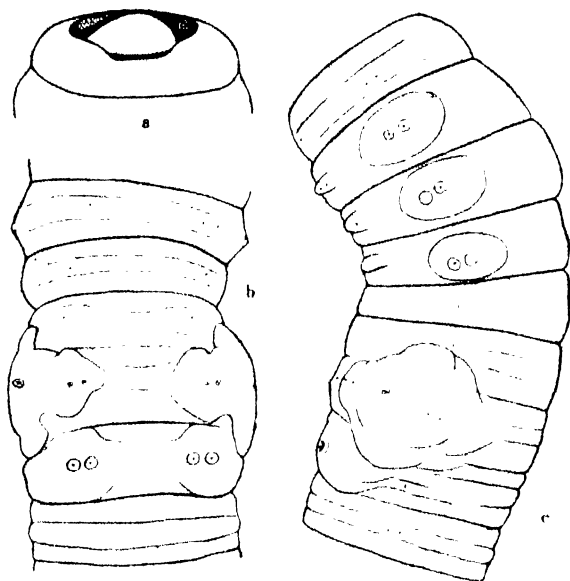


Fig. 4 *Allolobophora hataii*, n. sp. a, prostomium, ca. $\times 22.3$; b, ventral view of XII-XVII; c, lateral view of IX-XVII; b & c, ca. $\times 12.6$.

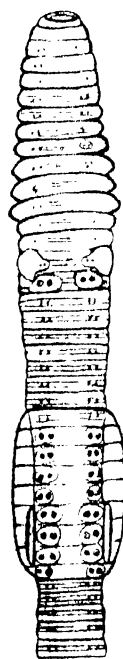


Fig. 5 *Allolobophora hataii*, n. sp. Ventral view of the anterior part of the body, drawn with free hand.

large sacs, each of somewhat triangular shape. These genital setae are almost similar in form to one another; almost straight but proximally slightly curved, grooved at nearly distal half, rather bluntly pointed at distal end; setae cd of X-XIII are slightly longer than the others, about 0.7 mm long.

Localities and materials: Chihnsien, 18 clitellate and 3 acitellate sps.; Tashihchiaio, 1 clitellate sp.

Remarks:

The specific name is dedicated to my teacher, Dr. SHINKISHI HATAI.

The present species closely resembles in many respects *All. prashadi* STEPHENSON which was recorded from India. But, it differs from the latter in the constant occurrence of the genital papillae on XV and XVI and in the aspect of the male pore (perhaps also in the characteristic features of the papillae of X-XII). No spermathecae were recognizable in the parietal wall.

16. *Allolobophora harbinensis*, n. sp.

(Figs. 6 & 7)

Description:

Length 76-96 mm, greatest diameter 2.7-3.3 mm (in the clitellar region), number of segments 134-144. Segments near the male pore region and those posterior to the clitellum are of 3 annuli; but, the annulation is generally indistinct. Colour uniformly grey except the clitellum which is brownish yellow.

Prostomium proepilobous. First dorsal pore in $4/5$ (in all specimens).

Setae closely paired; setal distance aa is nearly twice as ab; ab is slightly greater than cd; dd is less than $1/2$ of the circumference; aa : ab : bc : cd : dd : $1/2 \mu = 93 : 7.5 : 45 : 6 : 178 : 194$ on a segment immediately posterior to the clitellum. Setae ab of XXVII-XXXII are situated on papillae; ab of IX and XII and cd of IX, X and XII are also situated on papillae, but some of these are often absent (Fig. 6).

Clitellum saddle-shaped, extending from XXV or $\frac{1}{3}-\frac{1}{2}$ XXV or seldom XXVI, to XXXII or $\frac{1}{4}-\frac{1}{2}$ XXXIII ($=7-8\frac{1}{2}$). Pubertatis tubercles usually in XXIX-XXXI, or sometimes in XXIX- $\frac{1}{2}$ XXXI, narrow but distinct groove with slightly elevated margin, immediately lateral to the papillae in position (Fig. 6).

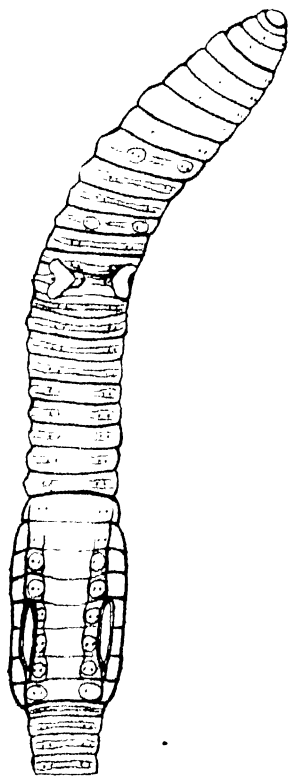


Fig. 6. *Allolobophora harbinensis*, n. sp. Ventral view of the anterior part of the body, drawn with free hand.

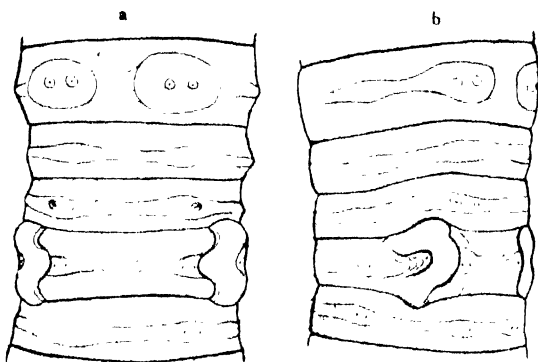


Fig. 7. *Allolobophora harbinensis*, n. sp. a, ventral view of XII-XVI; b, lateral view of XII-XVI; a & b, ca. $\times 12.6$.

On each side of XV, there is found a moderately elevated papilla, somewhat resembling a horse-shoe in shape: it is laterally opened, encroaching a very little into both of XIV and XVI, just lateral in position to ventral setae; male pore is found in the mid-area of the horse-shoe (Fig. 7, a & b).

Female pores are clearly visible on XIV, just lateral to seta b (Fig. 7, a & b).

Spermathecal pores, two pairs in 9/10 and 10/11, on cd-line.

Septa 6/7-8/9 moderately, 9/10 slightly thickened, the rest thin. Gizzard in XVII-XVIII.

Testes and funnels are free in X and XI. Seminal vesicles four pairs in IX-XII, darkened in colour; the posterior two pairs are larger than the anterior ones, and are nodular in appearance or form a few number of small spherical lobules; those in X are nearly equal in size to those in IX. Spermathecae small; ampulla spherical, thin-walled, with a relatively long duct; through the thin body wall they are usually seen externally.

The ventral and lateral setae of IX, X and XII are seen through the internal dissection to be contained in large and somewhat triangular sacs; they are proximally grooved and are about 0.8 mm long. The ventral setae of XXVII-XXXII are similarly grooved and are about 0.5 mm long, but these are contained within the usual setal sacs.

Locality and material: Harbin, 11 clitellate and 6 aclitellate specimens.

Remarks:

The present species is closely allied to *All. prashadi* and *All. hataii*, but differs from these mainly in having spermathecae and in the aspect of the male pore.

17. *Allolobophora dairenensis*, n. sp.

(Fig. 8 & 9)

Description:

Length 80-111 mm, greatest diameter 3.5-5.5 mm, number of segments 137-139. Except the anterior end, body dorsoventrally flattened and somewhat four-edges; behind the male pore region (except the clitellum) segments are tri-annular; both anterior and posterior ends are rather bluntly terminated. Colour uniformly pinkish; clitellum is fleshy red and resembles that of *Drawida gisti* MICHAELSEN.

Prostomium proepilobous. First dorsal pore in 4/5.

Setae closely paired; setal distance aa is nearly twice as bc; ab is slightly greater than cd; dd is smaller than $1/2$ of the circumference; aa : ab : bc : cd : dd : $1/2 \mu$ = 83 : 8 : 38 : 6 : 132 : 160 in a segment immediately posterior to the clitellum. Setae cd of X, XI and XII (or most of them), and ab of IX (on one side in one specimen), XV, XVI and of clitellar segments are situated on papillae (Fig. 8; Fig. 9, a & b).

Clitellum saddle-shaped, in XXIII-XXXIII (=11). Pubertatis tubercles in XXIX-XXXI distinct in outline being ridged by three prominent papillae (Fig. 8; Fig. 9, a).

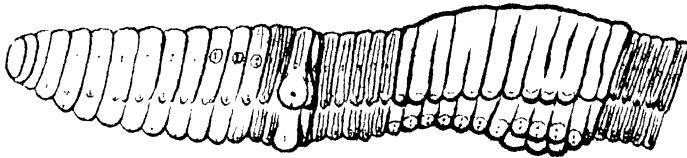


Fig. 8. *Allolobophora dairenensis*, n. sp. Lateral view of the anterior part of the body, drawn with free hand.

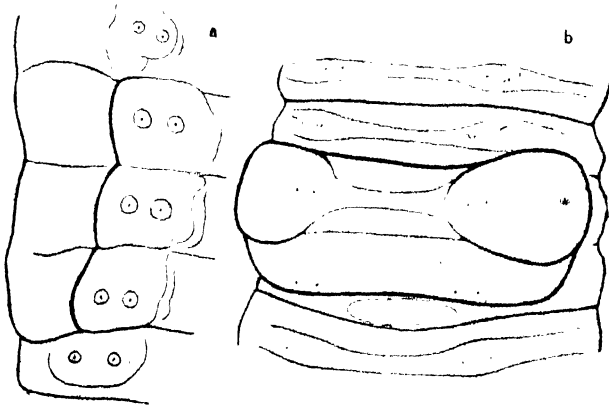


Fig. 9. *Allolobophora dairenensis*, n. sp. a, pubertatis tubercle; b, ventral view of XIII-XVII; a & b, ca. $\times 12.6$.

Male pores on XV; seated on prominent papillae, somewhat ovoidal, slightly encroaching into both of XIV and XVI; setae ab are planted on medial border of the male pore-papilla and cd are just dorsal to it. The ventral portion lying between the male pore-papillae and the ventral surface of the anterior two thirds of XVI are also elevated but in a slight degree. Thus, XV and the greater part of XVI appear to form, as a whole, an elevated rectangular plate. On XVI, the setae ab are planted on the posterior margin of the elevation (Fig. 9, b).

Female pores, one pair on XIV, just lateral to seta b (Fig. 9, b).

Spermathecal pores are absent.

Septa 5/6, 10/11 and 11/12 slightly thickened, 6/7-9/10 moderately thickened and muscled, the rest thin. Crop in XV-XVI; gizzard in XVII-XVIII.

Testes and funnels are free in X and XI. Seminal vesicles four pairs in IX-XII, whitish: the anterior two pairs are much smaller than the posterior ones; those in X are slightly smaller than those in IX.

Spermathecae are entirely absent.

The setae cd of IX-XII, and ab of IX, XV, XVI and of the clitellar segments are found through the internal dissection to be contained in large sacs. Except the proximal end which is slightly curved, they are almost straight, grooved and moderately pointed; those of IX-XII about 0.77 mm long, those of XVI about 0.6 mm long, those of the clitellar segments about 0.66 mm long and those of XV about 0.57 mm long. Those of XV are grooved at about distal 3/5-4/5, while the rest of the genital setae grooved at about distal half.

Locality and material: Dairen, 3 clitellate and 2 acitellate specimens.

Remarks:

Among the athecal species of the genus, the present species resembles *All. prashadi* and *All. hataii*, but differs from these mainly in the colouration of the body, in the body shape, in the extension of the clitellum, in the male pore aspect and in the shape of the pubertatis tubercle.

18. *Allolobophora jeholensis*, n. sp.

(Figs. 10 & 11)

Description:

Length 41-53 mm, greatest diameter 4-4.6 mm; number of segments 132-140. Body cylindrical in form; each segment behind the male pore region with 3 annuli. Unpigmented; body appears generally pale and clitellum limy white.

Prostomium, epilobous 1/4 1/3 or proepilobous (Fig. 10, a-c).

First dorsal pore in 4/5.

Setae closely paired; setal distance ab is larger than cd; aa is nearly twice as bc; dd is a little smaller than 1/2 of the circumference; aa:ab:bc:cd:dd=40:3.8:22:2.6:78 in a segment immediately posterior to the clitellum. The setae cd of IX-XII (or most of them) and ab of XV, XVI and of the clitellar segments are planted on papillae; the

papillae on the clitellar segments are less distinct than those on the rest.

Clitellum saddle-shaped, moderately swollen, in XXIII-XXXII or -XXXIII (=10 or 11). Pubertatis tubercle distinctly ridged; the region just lateral to the genital papillae of XXIX-XXXI is thickened and distinctly stretched lateralwards. The ventral part of these segments are also, however, a little thicker than the remaining part of the clitellum. Even in most of the acitellate specimens, the characteristic appearance of the clitellar region is slightly developed (Fig. 11).

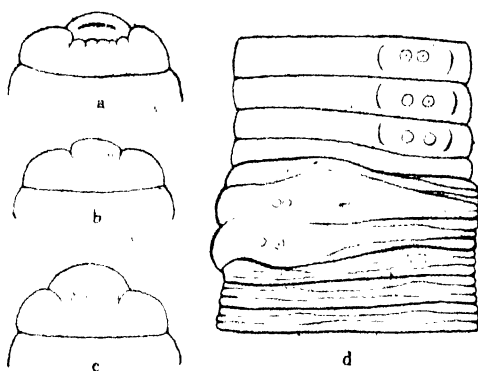


Fig. 10. *Allolobophora jeholensis*, n. sp. a-c, prostomium; d, lateral view of X-XIX; a-d, ca. $\times 12.6$.

Male pores are situated on XV, between b and c, slightly nearer to b; seated on prominent papillae of somewhat ovoidal shape, encroaching anteriorly into the greater part of XIV and posteriorly a very little into XVI (setae ab of XVI are planted on the elevation); the mid-ventral portion of XIV-XVI is also slightly elevated, thus the ventral part of these segments appears to be, as a whole, an elevated rectangular plate (Fig. 10, d).

Female pores, one pair on XIV, just lateral to b (Fig. 10, d).

Spermathecal pores are absent.

Septa 6/7-9/10 moderately thickened, the rest thin. Crop in XV-XVI; gizzard in XVII-XVIII.

Testes and funnels are free in X and XI. Seminal vesicles are four pairs in IX-XII, whitish; the anterior two pairs are smaller than the

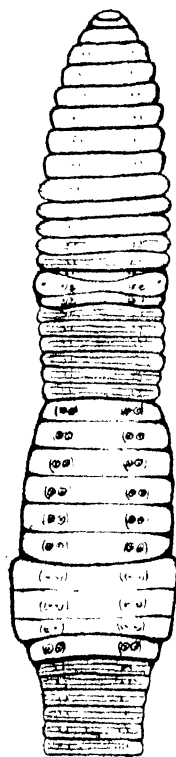


Fig. 11. *Allolobophora jeholensis*, n. sp. Ventral view of the anterior part of the body, drawn with free hand.

posterior ones; those in X are a little smaller than, or nearly equal to those in IX.

Spermathecae are entirely absent.

Performing an internal dissection, the setae *cd* and *ab* on the genital papillae are found to be contained within the respective large sac; sacs in IX-XII are larger than those in others. Setae *cd* in IX-XII about 0.84 mm long, *ab* in XV about 0.74 mm long, those in XVI and in the clitellar segments about 0.6 mm long. They are similar in shape to one another; almost straight except the proximal end which is slightly curved, and are grooved and moderately pointed distally.

Locality and material: Chihfen, 8 clitellate, 5 semi-clitellate and 14 a clitellate sps., September 18, '37, by Mrs. S. KOHASI and H. TADA.

Remarks:

The present species differs from the other athecal members of the genus mainly in the characteristic appearance of the clitellar region and of the male pore region.

19. *Allolobophora caliginosa* (SAV.) forma typica 1826

1924 *All. caliginosa typica*, SVETLOV, Bull. Inst. Rech. Biol. Perm, II, p. 323.

1926 *All. caliginosa typica*, PICKFORD, Nat. Hist. Wicken Fen, III, p. 228, figs. 4, A & B.

1931 *All. caliginosa typica*, MICHAELSEN, Zool. Jahrb. Syst., p. 537.

1934 *All. caliginosa typica*, SCIACCHITANO, Fauna Parco Naz. Gran Paradiso, III, p. 4.

1935 *All. caliginosa typica*, ČERNOSVITOV, Monogr. tschechosl. Lumb., pp. 52-53, fig. 35.

(Literature: see this paper).

1937 *All. caliginosa typica*, ČERNOSVITOV, Mitt. Königl. Naturwiss. Inst., X, p. 85.

1937 *All. caliginosa typica*, TERRY, Bull. Mus. Hist. Nat., IX, pp. 146-148.

Description:

Length 125-156 mm, greatest diameter 4.2-4.8 mm, number of segments 148-169. Colour uniformly light slate-blue. Prostomium, epilobous ca. 1/3. Most of the segments behind male pore region with 3 annuli which are rather indistinct; postclitellar region appears somewhat moniliform. First dorsal pore in 9/10.

Setae closely paired, $aa > bc$ and *dd* nearly equal to 1/2 of the circumference. Setae *ab* of IX-XI and of XXX, XXXII and XXXIV (or sometimes, of XXX and XXXII or XXXII and XXXIV) are planted on genital papillae. The genital papillae of IX-XI are not so prominent; those of the clitellar region are distinct but irregular in outline.

Male pores slit-like, between *b* and *c* in XV, seated on prominent papillae which encroach into both of XIV and XVI.

Spermathecal pores, two pairs in 9/10 and 10/11, on cd-line.

Clitellum saddle-shaped, usually in XXVII-XXXV or rarely in XXVII $\frac{1}{2}$ -XXXV (=8 $\frac{1}{2}$ -9). In most cases, pubertatis tubercles are rather intermediate in position and also in shape between f. *typica* and f. *trapezoides*. But, its shape and position differ from those in f. *trapezoides*; i. e. (1) "puberty wall" in this form consists of two elevations lateral to seta b in XXXI and XXXIII (on each side); (2) even if each elevation encroaches into both of the anterior and posterior segments, there are always found a transverse gap separating these two elevation from each other; (3) sometimes, in addition to the formers there is found an indistinct elevation in XXXII, but these three are never coalesced entirely with one another and thus never form a single glassy groove-like striation as in the case of f. *trapezoides*. In semi-clitellate specimens, the puberty walls are clearly recognized being restricted in XXXI and XXXIII.

Septa 5/6-11/12 moderately or much thickened: the following ones become gradually thinner. Seminal vesicles of the anterior pairs are smaller than the posterior ones. Spermathecae provided each with a stalk are small, embedded within the thickened septa. Setae ab on the genital papillae are found, through an internal dissection, to be contained within moderately large sacs: these setae are almost straight except the slightly curved proximal end, and are grooved and moderately pointed distally.

Locality and material: Harbin, 11 clitellate and 2 aclitellate specimens.

Distribution: Whole Europe, Siberia, N. India, N. & S. America, S. Africa, Azores, Madeira, Australia, New Zealand, Manchoukuo.

20. *Allolobophora caliginosa* (SAV.) forma *trapezoides* (A. DUG.) 1828

- 1926 *All. caliginosa trapezoides*, PICKFORD, Nat. Hist. Wicken Fen, III, p. 228, fig. 4, c.
 1931 *All. caliginosa trapezoides*, CHEN, Contr. Biol. Lab. Sci. Soc. China, Zool. Ser., VII, 3, pp. 168-169.
 1933 *All. caliginosa trapezoides*, CHEN, *ibid.*, IX, 6, pp. 216-217, figs. 11 & 12.
 1934 *All. caliginosa trapezoides*, SCIACCHITANO, Arch. Zool. Italiano, XX, p. 14.
 1935 *All. caliginosa trapezoides*, ČERNOSVITOV, Monogr. tschechosl. Lamb., p. 53. (Literature: see this paper).
 1935 *All. caliginosa trapezoides*, ČERNOSVITOV, Mem. Soc. Zool. Tscécosl. Prague, III, p. 4.
 1935 *All. caliginosa trapezoides*, KOBAYASHI, Zool. Magaz., 47, p. 130.
 1936 *All. caliginosa trapezoides*, NOMURA and KOBAYASHI, *ibid.*, 48, pp. 885-893.
 1937 *All. caliginosa trapezoides*, ČERNOSVITOV, Mitt. Königl. Naturwiss. Inst., X, p. 85.
 1937 *All. caliginosa trapezoides*, GAVRILOV, Zool. Anz., 118, p. 146.
 1938 *All. caliginosa trapezoides*, KOBAYASHI, Ann. Zool. Japon., XVII, 3-4, p. 414.
 1938 *All. caliginosa trapezoides*, KOBAYASHI, Jour. Chosen Nat. Hist. Soc., XXIV, pp. 6.

It is noteworthy that, except in the case of f. *trapezoides*, both of f. *typica* and f. *trapezoides* are rather small in number. The former subspecies is not found in the southern part of Manchoukuo, and the latter was not found in the northern. In Korea, the latter has been found in almost every locality where collections have been made (except in the far northern regions). Judging from their distribution in Manchoukuo, f. *typica* may be a "Kälteform" and f. *trapezoides* a "Wärmeform" as MICHAELSEN mentioned (10 a).

Localities and materials: Dairen, 8 clitellate and 9 acitellate sps.; Hunho near Fengtien, 1 clitellate sp.; Hushun near Fengtien, 3 clitellate sps.; (various parts of the Kwanto Province, based upon the verbal communication from Mr. J. ÔIZUMI).

Distribution: Widely distributed in the temperate zone: Europe, Transcaucasia, Turkey, India, Persia, Japan, China, Egypt, Algeria, S. Africa, Australia, N. & S. America, Manchoukuo.

21. *Allolobophora* sp.

Unfortunately as the specimens are of acitellate and already macerated, it was unable to identify the species definitely. Apparently they seem to differ from the other Lumbricid worms of Manchoukuo dealt with in the present paper. Body-size 140 × 5.5 mm. Prostomium, epilobous ca. 2/3; posterior demarcation distinct. First dorsal pore in 4/5. Setae closely paired, aa > bc. Spermathecal pores, two pairs in 9/10 and 10/11, on cd-line.

Locality and material: Tiehsi near Fengtien, 2 acitellate specimens, May, '37, by Mr. K. KOBAYASHI.

Genus *BIMASTUS* MOORE 1893

22. *Bimastus parvus* (EISEN) 1874

- 1933 *Bimastus parvus*, CHEN, Contr. Biol. Lab. Sci. Soc. China, Zool. Ser., IX, 6, pp. 222-224, fig. 13.
 1936 *Bimastus* sp., KOBAYASHI, Sci. Rep. Tôhoku Imp. Univ., Biol., XI, 1, p. 183.
 1937 *Bimastus parvus*, ČERNOSVITOV, Rec. Ind. Mus., XXXIX, 2, p. 111.
 1938 *Bimastus parvus*, GATES, Bull. Raff. Mus., Singapore, XIV, p. 222.
 1938 *Bimastus parvus*, KOBAYASHI, Jour. Chôsen Nat. Hist. Soc., XXIV, p. 6.

Description:

Length 31-72 mm, greatest diameter 1.2-2.7 mm (in clitellar region), number of segments 91-124. Colour, dorsally red and concentrated in

anterior part, clitellum grey or pinkish in some specimens with incomplete clitellar glandularity. Prostomium, epilobous ca. $1/2-4/5$. First dorsal pore in $5/6$.

Setae closely paired: setal distance $aa:ab:bc:cd:dd=18:4:15:3:48$ in a segment immediately posterior to the clitellum; dd is nearly equal to $1/2$ of the circumference.

Clitellum saddle-shaped, in XXIV-XXX ($=7$); in one Fengtien-specimen in XXIII $\frac{1}{2}$ -XXX ($=7\frac{1}{2}$), in one Mutanchiang-specimen in XXIII-XXX ($=8$). Pubertatis tubercles are always indistinct and often not recognizable; in XXV-XXVIII, or -XXIX, or -XXX.

Male pores on XV, each seated on a whitish, slight elevation, between b and c . Female pores on XIV, close and lateral to b .

No septa specially thickened. Seminal vesicles in XI and XII. Spermathecae absent.

Localities: Harbin, Chiamussu, Mutanchiang, Kirin, Yenki, Tumen, Hsinking, Chaoyangchen, Shanchengchen, Fengtien, Tashihchiao, Antung, Chihnsien.

Distribution: Japan, Korea, China, Tibet, India, St. Paul Island, S.W. Africa, N. & S. America, Java, Malay Peninsula, Manchoukuo.

23. *Bimastus beddardi* (MICHAELSEN) 1894

1900 *Helodrilus* (*Bimastus*) *beddardi*, MICHAELSEN, Tierreich, pp. 502-503.

1901 *Helodrilus* (*Bimastus*) *beddardi*, MICHAELSEN, Bull. Acad. Imp. Sci. St.-Petersburg, XV, 2, p. 213.

1902 *Helodrilus* (*Bimastus*) *beddardi*, MICHAELSEN, Jahrb. Hamb. Wiss. Anst., XIX, 2, p. 50.

1910 *Helodrilus* (*Bimastus*) *beddardi*, MICHAELSEN, Ann. Mus. Zool. Acad. Imp. Sci. St.-Petersburg, XV, p. 64.

1917 *Helodrilus* (*Bimastus*) *beddardi*, SMITH, Proc. U. S. Nat. Mus., LII, pp. 15-16.

Description:

Length 29-68 mm, greatest diameter 1-2.7 mm (in clitellar region), number of segments 97-125. Colouration similar to that of *B. parvus*. Prostomium, epilobous ca. $1/2-2/3$. First dorsal pore in $5/6$.

Setae closely paired: setal distance $aa:ab:bc:cd:dd=20:4:17:3:57$ in a segment immediately posterior to the clitellum; dd is nearly equal to $1/2$ of the circumference.

Clitellum saddle-shaped, in XXIV-XXXI ($=8$); out of 11 clitellate Harbin-specimens, in two cases in XXIV- $\frac{1}{2}$ XXXII ($=8\frac{1}{2}$) and in four cases in XXIII-XXXI ($=9$); out of 21 clitellate Chiamussu-specimens, in

two cases in XXIII-XXXI ($=9$); out of 21 clitellate Paichengtze-specimens, in one case in XXIV-XXXII ($=9$), in seven cases in XXIII-XXXI ($=9$) and in one case in XXIII $\frac{1}{2}$ -XXXI ($=8\frac{1}{2}$). Pubertatis tubercles are always indistinct, and sometimes are not recognizable.

Male pores on XV, each seated on a whitish, slight elevation, between b and c. Female pores on XIV, close and lateral to b.

No septa specially thickened. Seminal vesicles in XI and XII. Spermathecae absent.

Localities: Paichengtze, Keshan, Harbin, Chiamussu, Mutanchiang, Shanchengchen.

Distribution: N.-E. Mongolia, Tibet, Irland, N. America, Hawaiian Island, Manchoukuo.

A note on *B. parvus* and *B. beddardi*.

As stated in the foregoing descriptions, *B. parvus* and *B. beddardi* resemble each other. MICHAELSEN ('10 a, p. 64) has expressed a doubt concerning their specific distinction. SMITH ('17, p. 17) has stated that their resemblance is only apparent, and that they are actually distinct from each other. (And, the same writer ('27) asserts that the structures of the calciferous glands of both species conform with that of *B. gieseleri*.) According to the study he has made of the American specimens, *B. beddardi* differs from *B. parvus* in the following points: (1) the clitellum uniformly extends a little beyond the posteriad; (2) the body is somewhat longer, but the number of segments is less; (3) the setae are more closely paired; (4) the anterior segments are comparatively a little smaller than the others, and the prostomium is broad and blunt, which all tend to give the anterior a comparatively broad and blunt appearance (in *B. parvus*, the anterior segments are decidedly more reduced in diameter, and the end seems to be considerably pointed).

A careful examination was made of these four characteristics.

(1) Clitellum. In *B. beddardi*, usually it extends from XXIV-XXXI, and in one specimen into XXXII and in two specimens into $\frac{1}{3}$ XXXII; while in *B. parvus* usually in XXIV-XXX. In both species, it is not unusual for the clitellum to begin to swell in XXIII. MICHAELSEN ('09 & '10 a) has also reported on similar cases in the Kashmir- and the Chinese-specimens of *B. parvus*, and of the Tibetan specimens of *B. beddardi*. In a few cases of both Chiamussu- and Tumen-specimens, the clitellum was seen extending from XXIV or XXIII $\frac{1}{2}$ - $\frac{1}{3}$ XXXI or $-\frac{1}{2}$ XXXI.

If no further distinct differences are recognized in other characteristics, in which species should these specimens be classified? And also, in many specimens of *B. parvus* the clitellum invades, though very slightly, into XXXI. But, if the setal ratio is taken into consideration, it becomes clear that such ambiguous specimens must belong to *B. parvus*.

(2) Body length and number of segments. In *B. parvus*, the body length is 31-72 mm and the number of segments 91-124; while in *B. beddardi*, the former is 29-68 mm and the latter is 97-125. Thus, both species closely resemble each other in these characteristics. In the case

Body length (mm.)	No. of specimens		No. of segments	No. of specimens	
	<i>parvus</i>	<i>beddardi</i>		<i>parvus</i>	<i>beddardi</i>
29	0	1	91	1	0
30	0	1	92	0	0
31	1	0	93	0	0
32	1	0	94	0	0
33	3	1	95	0	0
34	1	0	96	2	0
35	1	0	97	0	1
36	1	1	98	0	0
37	2	2	99	1	2
38	2	1	100	0	1
39	2	2	101	1	0
40	2	3	102	1	2
41	1	0	103	0	2
42	1	2	104	1	1
43	1	5	105	3	1
44	2	1	106	0	2
45	2	5	107	1	1
46	2	2	108	3	0
47	2	4	109	2	0
48	1	3	110	0	2
49	3	2	111	1	3
50	3	3	112	2	2
51	2	1	113	3	4
52	1	2	114	4	4
53	3	2	115	5	0
54	2	1	116	3	5
55	2	3	117	5	5
56	0	0	118	2	2
57	1	1	119	0	2
58	0	0	120	2	1
59	0	0	121	0	2
60	0	1	122	1	0
61	1	1	123	1	1
62	0	1	124	2	0
63	0	0	125	0	1
64	0	0			
65	0	1		47	47
66	0	0			
67	0	0			
68	0	1			
72	1	0			
	47	54			

of *B. parvus*, the largest number of segments hitherto known is 111 as reported by SMITH ('17) of the specimens taken from America and it is also 111 as reported by CHEN ('31 & '33) of the specimens obtained from Central China. In the case of *B. beddardi* it is 97 as reported by SMITH of the specimens which came from America. In the Manchoukuo specimens, the number of segments in both species is considerably larger than those reported in both the American and Chinese specimens. The body length and the number of segments are of systematic importance, and they show a slight or sometimes a considerable variation. Unless the extent of the variation of these characteristics is clearly recorded, we shall not be able to estimate their significance. But in the Manchoukuo specimens of these two species, though they are rather few in number, no marked difference in these characteristics was found. The frequency of the body length and number of segments is indicated above.

(3) Setal distance. In several specimens, the setal distance was measured from one segment situated immediately posterior to the clitellum. The results are indicated below. In the Manchoukuo specimens, the setal ratio in *B. beddardi* is very little larger than that in *B. parvus*; such

Setal distance & localities	aa:ab:bc:cd:dd	Localities
Species		
<i>B. parvus</i>	15: 4:13: 3:43	Harbin
	17: 4:15: 3:53	Chiamussu
	20: 4:17: 3:50	Mutanchiang
	18: 4:14: 3:43	Chaoyangchen
	21: 4:16: 3:53	Fengtien
	16: 4:14: 3:40	Yenki
	20: 4:17: 3:53	Tumen
Mean value of setal distance	18: 4:15: 3:48	
	18: 4:16: 3:48	N. America, by SMITH
<i>B. beddardi</i>	19: 4:16: 3:54	Keshan
	20: 4:17: 3:58	Paichengtze
	21: 4:18: 3:56	Harbin
	18: 4:16: 3:57	Chiamussu
	21: 4:17: 3:59	Mutanchiang
Mean value of setal distance	20: 4:17: 3:57	
	26: 4:20: 3:80	N. America, by SMITH

distinct difference as seen in the American specimens are not to be perceived. Besides, the setal ratio obtained of the Fengtien-, Tumen- and Mutanchiang-specimens in the case of *B. parvus* is nearly equal to that of *B. beddardi*. On account of such trifling difference in the setal ratio, it is very difficult to identify the species. And, as already stated in the case of the clitellum (1), identification may be hardly possible at all if the clitellar extension is taken into consideration together with the setal ratio too.

(4) Shape of the anterior end of the body. No constant difference between the two species was found in the shape of the anterior end of the body and in the shape of the prostomium.

In the opinion of SMITH, with whom on this point I agree, the close resemblance of *B. parvus* and *B. beddardi* is only apparent the two being really distinct from each other. The distribution of each species appears to endorse this conception. But, at the same time it seems to be reasonable to treat *B. beddardi* as a variety of *B. parvus*. Thus, the re-examination of the original specimens and the close study of the variations in the important features of the American specimens of these two species seem to be very desirable.

Genus OCTOLASIMUM OERLEY 1885

24. *Octolasion lacteum* (OERLEY) 1885

(Fig. 12)

1924 *Octolasion lacteum*, SVETLOV, Bull. Inst. Rech. Biol. Perm., 2, pp. 324-325.

1932 *Octolasion lacteum*, SCIACCHITANO, Parco Naz. Gran Paradiso, III, p. 6.

1934 *Octolasion lacteum*, SCIACCHITANO, Arch. Zool. Italiano, XX, p. 18.

1935 *Octolasion lacteum*, ČERNOSVITOV, Monogr. tschechosl. Lumb., pp. 70-71, figs. 59-60. (Literature: see this paper).

1936 *Octolasion lacteum*, ČERNOSVITOV, Rec. Ind. Mus., XXXIX, 2, p. 111.

1937 *Octolasion lacteum*, ČERNOSVITOV, Mitt. Königl. Naturwiss. Inst., X, p. 89.

1938 *Octolasion lacteum*, ČERNOSVITOV, Zool. Anz., 122, p. 288; 123, p. 199.

Description:

Length 131 mm, greatest diameter 4 mm, number of segments 126. Body cylindrical; secondary annulation indistinct or may be said to be absent. Colour in formalin, pinkish grey; unpigmented; clitellum light flesh.

Prostomium, epilobous ca. 1/2. First functional dorsal pore in 12/13, distinct; in 10/11 and 11/12 indistinct and non-functional pores are found.

Setae found on segments anterior to male pores are paired, ab<bc>cd

or $aa:ab:bc:cd:dd:1/2\mu=29:6:16:4:108:95$ in IX; those posterior to male pores are either widely paired or separated, $ab>bc>cd$ or $aa:ab:bc:cd:dd:1/2\mu=27:11:10:7:63:73$ in a segment immediately posterior to the clitellum. Setae a and b of XII (left side only) are planted on a small but distinct genital papilla; on internal dissection they are found to be contained within usual setal sac; about 0.7 mm long, grooved at about distal half, nearly straight but proximally slightly curved, distally pointed rather bluntly.

Clitellum saddle-shaped, in XXX-XXXV (=6). Pubertatis tubercles in $\frac{1}{3}$ XXX- $\frac{1}{3}$ XXXV, darkened, each of a long groove (Fig. 12).

Male pores slit-like in XV, between b and c, nearer to b; each on a marked elevation which encroaches a little into the both of XIV and XVI (Fig. 12).

Female pores on XIV, just lateral to seta b.

Spermathecal pores, two pairs in 9/10 and 10/11, on c-line.

Any septa not thickened especially; 6/7-8/9 moderately thickened.

Seminal vesicles, four pairs in IX-XII; those in IX and X are digitiform and clearly differ in shape from those in XI and XII. Testis sacs present. Spermathecae spherical, each with a very inconspicuous stalk.

Locality and material: Harbin, a single clitellate specimen.

Distribution: Widely distributed in Europe, Austria, Hungary, Yugoslavia, Bulgaria, Switzerland, Spain, France, Germany, England, Tschechoslovakia, Rumania, Italy, W. & S. Russia, Algeria, Azores, Canary Islands, Åland Islands, North America, Mexico, Uruguay, Australia, North India, Manchoukuo.

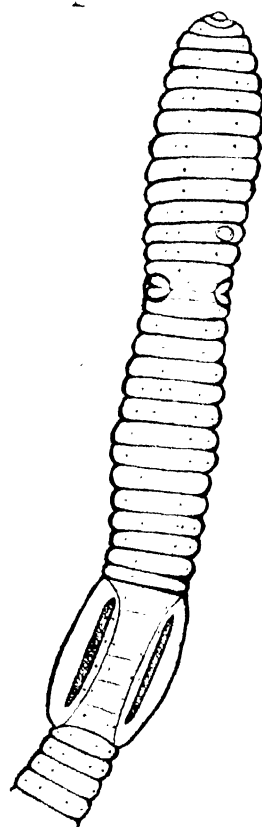


Fig. 12. *Octolasion lacteum* OERLEY. Ventral view of the anterior part of the body, drawn with free hand.

GENERAL DISTRIBUTION, AND THE AMOUNT OF RAINFALL
AND THE TEMPERATURE TAKEN AS THE DELIMITING
FACTORS FOR THE DISTRIBUTION OF THE
TERRESTRIAL OLIGOCHAETES

Manchōukuo is bounded on the west by the steppes and deserts of Gobi, and on the north by the severely cold plain of Siberia, while on the south and south-east it is bounded by North China, the Yellow Sea and Korea which are relatively temperate in climate. Thus, the worms in this country can be said to live under three different climatic environments, viz. aridity, low temperature and relatively moderate temperature. It is clearly recognizable from the present study that the amount of rainfall and the temperature are the most important factors in delimiting the distribution of the terrestrial oligochaetes.

The amount of annual rainfall and the annual mean temperature are illustrated in Fig. 13. As is clearly shown in the figure, the amount of annual rainfall gradually diminishes from east to west. The district

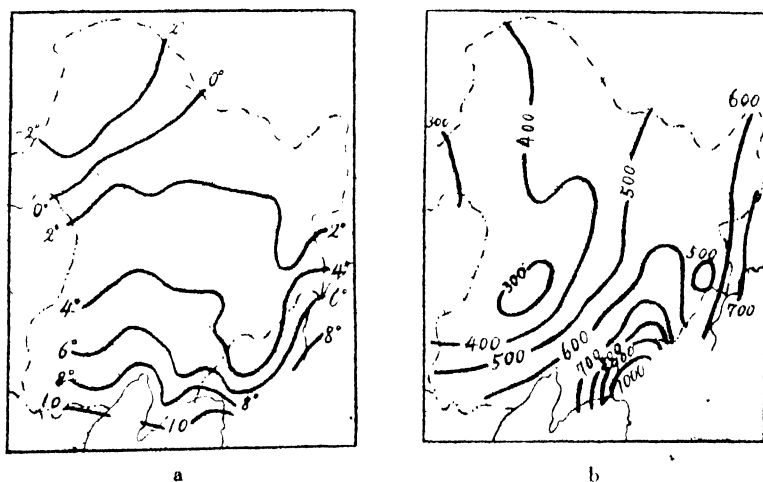


Fig. 13. a, annual mean temperature: b, amount of the annual rainfall (mm).
From OGA (31) (after TOBITA and IRIÉ). In every region, the monthly amount of rainfall is largest in summer and is smallest in winter.

which is situated to the west of the Manchurian Railway, is called "Eastern Gobi", which consists of alkaline soils, sandy lands and steppes, and in which the amount of the annual rainfall is less than 500 mm. In Mongolia it is probable that the amount of annual rainfall is only 200-350 mm, though no accurate measurements in that country have been

made. On the contrary, in the south-eastern district of Manchoukuo which lies close to Korea it amounts to as much as 1,000 mm.

In Manchoukuo the temperature becomes gradually lower towards the north and in the region north of the former Eastern Chinese Railway the annual mean temperature is lower than 2–3°C. But, in the coastal region and in the districts close to North China, it is about 8°–10°C.

By the 500 mm isohyet, Manchoukuo is divided into two parts, viz. the west which is arid and the east which is wet. By the two isothermal lines of 2°–3°C and of 8°C, this country may be again divided into three parts, viz. the north which is severely cold, the central cold and the south relatively temperate. Thus, if these two factors of rainfall and temperature are combined, Manchoukuo may be divided into six districts. They are the north-west (N.W.) arid and severely cold, the north-east (N.E.) wet and severely cold, the central-west (C.W.) arid and cold, the central-east (C.E.) wet and cold, the south-west (S.W.) arid and temperate, and the south-east (S.E.) wet and temperate. The distribution of the worms among these districts is illustrated in Fig. 15.

In the N.W. and C.W. districts where the amount of annual rainfall is less than 400–500 mm, no endemic species are found and the population of worms is generally very low. In the N.W., in the localities of Hailar, Ilkete and Tsitsihar, only a Siberian Lumbricid form, *Eisenia nordenskiöldi typica*, was collected.

In the C.W., four peregrine forms, viz. *Eisenia rosea macedonica*, *Allolobophora caliginosa typica*, *Bimastus beddardi* and *Drawida japo-*

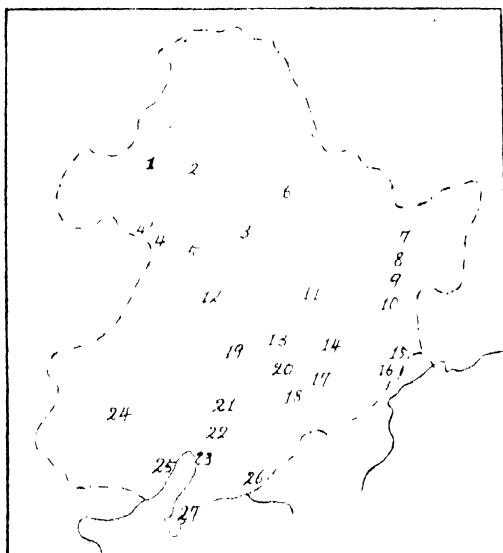


Fig. 14. Localities where the collections were made. 1. Hailar. 2. Ilkete. 3. Tsitsihar. 4. Nankiang. 4'. Halun-Arshan. 5. Wangvehmiao. 6. Keshan. 7. Chiamussu. 8. Iyasaka. 9. Poli. 10. Mutanchiang. 11. Harbin. 12. Paichengtze (Taoan). 13. Hsinking. 14. Kirin. 15. Tumen. 16. Yenki. 17. Chaoyangchen. 18. Shanchengchen. 19. Santaitze. 20. Fsi Feng. 21. Fengtien. 22. Anshan. 23. Tashihchiao. 24. Chihfen. 25. Chinhsien. 26. Antung. 27. Dairen.

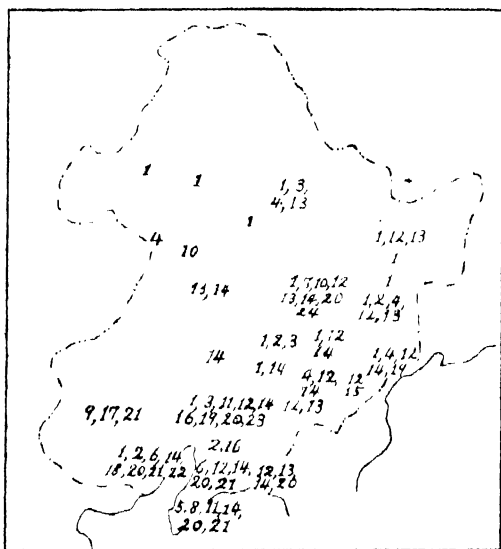


Fig. 15. Distribution of 24 forms in Manchoukuo.

1. *Eisenia nordenskiöldi typica*. 2. *E. nodenskiöldi manshurica*. 3. *E. rosea typica*. 4. *E. rosea macedonica*. 5. *E. foetida*. 6. *Allolobophora hataii*. 7. *All. harbinensis*. 8. *All. daurenensis*. 9. *All. jeholensis*. 10. *All. caliginosa typica*. 11. *All. caliginosa trapezoides*. 12. *Bimastus parvus*. 13. *B. beddardi*. 14. *Drawida japonica*. 15. *Dr. propatula*. 16. *Dr. koreana*. 17. *Dr. jeholensis*. 18. *Dr. gisti*. 19. *Dr. nemora*. 20. *Pheretima hupeiensis*. 21. *Ph. aggera*. 22. *Ph. tschiliensis*. 23. *All. sp.* 24. *Octolasion lacteum*.

districts of N.W. and C.W., but its fauna is clearly different from those of these two. The population of worms is higher in the N.E. district, and therefore it is not so difficult to collect them here as in the N.W. and C.W. For instance, as already stated, in Tsitsihar (N.W.) merely a single subspecies of *E. nordenskiöldi typica* was found, while in Keshan that is in a locality only about 170 kilometers distant in the north-east, the population of worms was higher, and four forms of *E. nordenskiöldi typica*, *E. rosea typica*, *E. rosea macedonica* and *B. beddardi* were found, though they were all peregrines. Further, in Mutanchiang was found an endemic form, *E. nordenskiöldi manshurica*, n. subsp. Possibly, like *f. lagodechiensis* form Transcaucasia it may be a southern form of this species.

The district C.E. mainly occupies the mountainous region, continuing

nica were found. The population of worms becomes gradually lower towards the west, and in such regions close to the political boundary of Mongolia as Nanhkingan and Halun-Arshan it was very difficult to find even a single specimen. (In spite of intensive endeavours made by myself and by coolies in searching in Halun-Arshan not a single worm was found, and in Nanhkingan only one specimen, i. e. *E. rosea macedonica*, was obtained.)

Dr. japonica was found with great difficulty in Paichengtze (=Taoan) which is located relatively close to the Manchurian Railway. This species was found to be most predominant in the wet spots of cultivated land.

The N.E. district is subequal in latitude to the two

eastwards to the north highland of the Korean peninsula. Although the amount of annual rainfall is more than 500 mm, the temperature is relatively low. The narrow plain-region along the Manchurian Railway, where the towns are scattered, is slightly higher in temperature than the mountainous region. In the towns distributed in this plain-region, such as Harbin and Fengtien, *Pheretima hupeiensis* was found. But, in the mountainous region it was not found (nor in the north highland of Korea). Excepting for the special occurrence of *Ph. hupeiensis*, the fauna of the C.E. district is represented by two groups of the genus *Drawida* and the family Lumbricidae. Two *Drawidian* forms of *Dr. nemora* and *Dr. koreana* appear to be widely distributed in this district; the first species was found in uncultivated land and the latter in both of cultivated and uncultivated land. *Dr. japonica* is the species mostly predominant in the cultivated land. It is noteworthy that *Dr. propatula* was found in Yenki, its original locality being Central China. A single specimen of *Octolasion lacteum* was obtained in Harbin; and the present report forms the first record concerning the existence of this species in Eastern Asia (excepting India). Eleven Lumbricid forms including two endemic ones were also found in this district. It means that the climate of this district is more favourable for the worms of this group than that of the other three districts above mentioned.

The survey tried in the S.W. district is very incomplete, and only a small number of specimens were collected from Chihfen, Jehol. This district occupies the mountainous region of Jehol, continuing westwards to Mongolia and southwards to the plain of North China. The greater part of Jehol is at the present day a treeless mountainous region, but it is well-known that, until the end of the Hshing dynasty, this land was covered by dense forests. According to the phytocological study of TAKAHASHI ('36), the amount of annual rainfall is only 200–300 mm, and the annual mean temperature is 5°–6°C in Chihfen where the worms were collected. (Both of these data are lower than those illustrated in Fig. 13.) From these data, we know that the climatic condition in Chihfen resembles, at least in the present day, that of Mongolia. Consequently, judging from the present environmental condition of Jehol it is surprising that three endemic species viz. *Ph. aggera*, *Dr. jeholensis* and *All. jeholensis* should be found there. Although the population of worms is very low, the conditions influencing the fauna of the S.W. district are similar to those in the S.E. district where the climatic condition is the most favourable in Manchoukuo for the existence of worms. The reason for

its being the most favourable in this respect is that the amount of annual rainfall is more than 600 mm, and the temperature higher than 8°C. Out of the 11 endemic species of Manchoukuo, 9 are found in this S.E. district. Except the special occurrence of *Ph. aggera* in Jehol and of *Ph. hupeiensis* in the plain-region of the C.E., the *Pheretima* species though few in number are almost entirely restricted to this S.E. district. Generally speaking, the members of the genus *Pheretima* appear to be adversely affected by a low temperature. Most of the endemic Lumbricid forms are found in this district, and this proves that even the worms belonging to the palaearctic group favour a relatively temperate climatic environment. A single specimen of *Dr. gisti*, which has been recorded from both of North China and Central Korea, was also found.

When summarizing the above statements, it is shown that: --

Rainfall and temperature are the most important factors in delimiting the distribution of the terrestrial oligochaetes. Aridity appears to be an absolute factor prohibiting their distribution. In the region where the amount of annual rainfall is less than 400 mm, no endemic species can exist. This hypothesis agrees with that of PICKFORD ('37) expressed in the study of Acanthodrilinae in South Africa. The effect of the degree of temperature, acting as a delimiting factor for the distribution of earthworms, appears to vary slightly according to the family, or the genus, or even to the species. An annual mean temperature higher than 8°C appears to be favourable for the existence of the terrestrial oligochaetes. This hypothesis seems to be applicable also to the causes of the distribution of the terrestrial oligochaetes throughout the world. The area in Manchoukuo which possesses such favourable climatic conditions as an amount of annual rainfall more than 400 mm and an annual mean temperature higher than 8°C, is confined only to the south coastal region. Consequently, the fauna of Manchoukuo generally is very poor in the number of worms, and at the same time, most of them are concentrated in this south coastal region. That some relics are found in the apparently unfavourable regions, may mean that the oligochaete fauna of this country was richer in geological times than at the present day. Such an idea appears to be endorsed by the occurrence of the endemic species of the genus *Drawida*, which constitutes phylogenetically an old group.

pH OF THE SOIL IN THE HABITATS

The soil-types are correlated both to the climate and the vegetation. Thus, if the soil-type affects the distribution of worms, it must be a factor

of secondary meaning. The soil samples, of which the degree of pH was measured, are only 29 in number.

pH of the soil in the habitats varied within the range of 5.2-7.4. Though it is well-known that a highly alkaline soil is found in the Eastern Gobi, soil samples of a high degree of pH in which worms lived, were not obtainable. In the following table the frequency of the occurrence of the worms is indicated corresponding to each pH value.

pH	No. of localities	Frequency of worms
5.2	1	±
5.4	1	±
5.6	1	++
5.8	2	##
6.0	0	
6.2	3	—; +
6.4	1	+
6.6	2	+
6.8	8	+; ++; ##
7.0	4	+; ##
7.2	5	+; ++
7.4	1	±

— means absent or very rare; ± present but rare; + little; ++ usual; ## many or relatively many.

From the above table it is recognizable that the soil in the range of 5.6-7.2 of pH appears to be favourable for the existence of worms. But, no worms were found in one case in which pH was 6.2. (This locality was in Halun-Arshan, where there is a hot-spring on the steppe, close to the political boundary of Mongolia and just west of the Hkingan Mts.)

It is rare that under natural conditions worms are found in the soil of pH 5 (see: for instance, NOMURA & KOBAYASHI, '36). Several worms of *E. nordenskiöldi typica* were found in the soil of pH 5.2 at Ilikete in the Hkingan Mts., in a spot at a short distance from a very small pasture, the worms being found only within the roots of plants. Their occurrence under such unfavourable conditions is noticeable, and we recognized that this species has a high degree of adaptability of a wide range of soil reaction, as well as to a low temperature. CHERYRKINA ('30) has also reported a similar case of the special adaptability of this species, in his study of the distribution of the Lumbricidae in the Ural region.

NORTHERN BOUNDARIES OF THE DISTRIBUTION OF BOTH
GENERA *PHERETIMA* AND *DRAWIDA*I. *Pheretima*.

In 1903 MICHAELSEN produced a map illustrating the distribution of the genus *Pheretima*. In this map both Manchoukuo and Korea are placed outside the region where *Pheretima* may be found. In his more recent maps ('31 & '34) both these countries are included in this region, an inclusion which is merely presumption on his part, and which is inaccurate as far as the distribution of the genus *Pheretima* in Eastern Asia is concerned.

In Tashihchiao, Chihnsien and in several localities of Kwanto Province, *Ph. aggera* was abundantly found, and in Chihfen, several juvenile specimens of the same species (?) were collected. In Korea, it is common in the region south of Kôkai-dô, and is abundant generally on the west side of the peninsula. In spite of my five years' search in Heian-hôku-dô, I was unable to collect even a single specimen of this species. From this fact, it may be concluded that Central Korea is rather more closely allied to the Liatung Peninsula than to the far west-northern part of Korea from the point of view of the distribution of earthworms. The distribution of *Drawida gisti* also indicates a similar relationship. In Chihnsien, in addition to *Ph. aggera*, *Ph. tschiliensis* was abundantly found. But, in Kaupangtze situated slightly north of Chihnsien, no endemic *Pheretima* species were found. Furthermore, according to the natives in this village, no large earthworms have been found here. *Ph. tschiliensis* is distributed on the west-side of Korea and also in North and Central China. Thus, no real endemic *Pheretima* species is found in Manchoukuo. If *Ph. aggera* may be considered to be a semi-endemic species, the northern boundary of the *Pheretima*-region should be marked in Manchoukuo by a line passing through Chihfen (?), Chihnsien and Tashihchiao. In Korea, *Ph. aggera* is found in Heijô, which is the northern limit of this species, and *Ph. sp.* (an endemic species) is found on Mt. Kongô. The region south of the line passing through these two localities belongs to the *Pheretima*-region (KOBAYASHI, '38). According to OHFUCHI ('38), in the North-eastern part of Japan proper, the endemic species are said to be merely about a dozen in number. Although the survey in Hokkaidô is as yet incomplete, the endemic species, *Ph. sp.* (the 5th species) is known to exist in Sapporo (YAMAGUCHI, '30) and *Ph. yezoensis* in Hakodate (KOBAYASHI, '38). The northern boundary in Hokkaidô may

possibly be drawn passing through Sapporo parallel to the line of latitude. When the survey in Hokkaidô is completed, the northern boundary of the *Pheretima*-region will become quite clear, and only the western boundary (in China) will then remain unknown.

II. *Drawida*.

Since the publication of the two reports, one by HATAI on *Dr. hatamimizu* from Japan ('30) and the other by MICHAELSEN on *Dr. gisti* from North China ('31), about a dozen endemic species have been recorded from Eastern Asia, a district which lies far distant from the centre of the genus of the present-day. Recently ('38) *Dr. gisti* was recorded by myself from Central Korea. It was included among materials from Chihnsien but was represented by a single specimen only. *Dr. nemora* is common in the far northern highlands of Korea (KOBAYASHI, '38), and *Dr. koreana* was reported by myself in the same paper the specimens being obtained from both Central and North Korea. These two species are distributed also in the central-eastern district of Manchoukuo. *Dr. jeholensis*, n. sp. was found in Chihfen, Jehol. A number of specimens of *Dr. propatula* were obtained in Yenki but were restricted to a single locality; this species has been recorded by GATES ('35) from Central China. Thus, four endemic species, *Dr. gisti*, *Dr. nemora*, *Dr. koreana* and *Dr. jeholensis* are known to occur in this country. Hsinking (ca. 43°55' N. L.) is a locality which corresponds to the northern limit in the *Drawida*-region; the northern boundary of the genus is approximately marked by the line passing through Chihfên Hsinking—Tumen. The northern boundary in Japan is not yet determined: no *Drawidian* forms have hitherto been reported from Hokkaidô. Recently, from the North-eastern part of Japan proper an endemic species, *Dr. moriokaensis* was reported by OHFUCHI ('38)*. Perhaps, this part may form the northern boundary in Japan proper.

It is very interesting to know that most of the endemic *Drawidian* species are found outside the *Pheretima*-region (cf. the distribution of the genus in India, Korea and Manchoukuo).

DISTRIBUTION OF THE FAMILY LUMBRICIDAE

The distribution of the family in both Europe and America has been made clear by many writers. But, that in Asia has been explained by

* Judging from his descriptions and illustrations, I think that his *Dr. ofunatoensis* seems to be synonymous with *Dr. nemora* and his *Dr. tatraensis* with *Dr. anchingiana*.

MICHAELSEN ('03 & '31) and by CERNOSVITOV ('32 & '35) basing their presumptions upon the general status of distribution. The presumption is that the large gap lying between Turkestan (or Tien-Shan or Kashmir and Punjab) and Japan may be connected by any of the courses (existence of endemic species) passing through (1) Siberia, (2) Mongolia or (3) China proper.

(1) Siberia.

It is much to be regretted that as the survey of the terrestrial forms found in the Baikal region is yet incomplete we are unable to compare its fauna with that of Manchoukuo. In North Manchoukuo no endemic worms are found. In the region close to the Transbaikal region only *E. nordenskiöldi typica* was found; and even in the region close to Ussuri, where the amount of rainfall is relatively abundant, only a few peregrine forms are found. And, even this Ussuri district (N.E.) appears to be more favourable for the existence of terrestrial worms than either the Baikal or Kamtschatka regions (MICHAELSEN, '00, '01, '10 a & '29). According to the study of CHETYRKINA ('30) even in the Ural region, which is, as is well-known, more favourable in climate for the distribution of plants and animals than Eastern Siberia, no endemic worms were found, and *E. nordenskiöldi typica* was most predominant, as in the case of North-eastern Manchoukuo. From the general status of the oligochaete fauna in South Siberia including North Manchoukuo, it is probable that no endemic Lumbricid worms are found there.

(2) Mongolia.

As already stated with regard to Manchoukuo on account of the climate, so also no endemic worms may possibly be found in Mongolia. It is improbable that any oasis-like area where worms might be found have remained free from the effects of the long-period-aridity. But, in a special locality such as Jehol (see: part of general distribution) some prearid endemic species may with difficulty have maintained their specific existence.

(3) China proper.

In spite of the fact that the survey of worms in China proper has recently been well executed, merely some peregrine forms such as *Bimastus beddardi*, *B. parvus* and *Allolobophora caliginosa trapezoides* have been reported. Even when any further researches are made, possibly no endemic Lumbricid species will be found in China except in North China.

Five endemic Lumbricid species are found in South and Central Manchoukuo. This fact means that even for these palaearctic worms a

relatively temperate climate is favourable, and that these districts are an important area in the Lumbricid zone in Asia. Therefore, the Lumbricid zone in Asia may be completed by a line passing through Turkestan (or Tien-Shan or Kashmir and Punjab)—the zonal area along the Great Wall of China (including the period of prearidity)—South Manchoukuo—Korea—Japan proper—Hokkaidô. The distribution of Lumbricidae given in the ČERNOSVITOV's map ('35, p. 22, Abb. 11) approximately agrees with that shown in the above statement.

REFERENCES

- BERG, L. S. 1925. Die Fauna des Baikalsees und ihre Herkunft. Arch. Hydrobiol., IV.
- ČERNOSVITOV, L. 1932. Die Oligochaetenfauna der Karpathen. II. Die Lumbricidae und ihre Verbreitung. Zool. Jahrb. Syst., LXII.
- 1935. Monographie der tschechoslovakischen Lumbriciden. Arch. Přír. vygk. Čech., XIX, 1.
- 1935 a. Oligochaeten aus dem tropischen Süd-Amerika. Capita Zool., VI, 1.
- 1935 b. Résultats zoologique du voyage de Mr. le Dr. J. STORKAN au Mexique. V. partie—Oligochaeta. Mem. Soc. Zool. Tchécosl. Prague, III.
- 1937. On a Collection of Indian Earthworms of the Family Lumbricidae. Rec. Ind. Mus., XXXIX, 2.
- 1937 a. Die Oligochaetenfauna Bulgariens. Mitt. Königl. Naturwiss. Inst., X.
- 1938. Zur Kenntnis der Oligochaetenfauna des Balkans. Zool. Anz., CXXII & CXXIII.
- CHEN, Y. 1930. On Some New Earthworms from Nanking, China. Sci. Rep. Nat. Cent. Univ., I, 1.
- 1931. On the Terrestrial Oligochaeta from Szechuan, with Descriptions of Some New Forms. Contr. Biol. Lab. Sci. Soc. China, Zool. Ser., VII, 3.
- 1933. A Preliminary Survey of the Earthworms of the Lower Yangtze Valley. *ibid.*, IX, 6.
- 1936. On the Terrestrial Oligochaeta from Szechuan. II., with the Notes on GATES' Types. *ibid.*, XI, 8.
- CHETYRKINA, I. 1930. Distribution of Lumbricidae in the Soils of the Troitsk District, Ural Region. Trav. Inst. Rech. Biol. et Stat. Biol. Univ. Perm., II. (after Biological Abstract).
- GATES, G. E. 1935. New Earthworms from China, with Notes on the Synonymy of some Chinese Species of *Drawida* and *Pheretima*. Smithon. Misc. Coll., XCIII, 3.
- 1935 a. On some Chinese Earthworms. Lingnan Sci. Jour., XIV, 3.
- 1938. Earthworms from the Malay Peninsula. Bull. Raff. Mus., Singapore, XIV.
- HATAI, S. 1930. On *Drawida hattamimizu*, Sp. Nov. Sci. Rep. Tôhoku Imp. Univ., Biol., V, 3.
- HIRAI, K., YOSHIDA, S. and FURUKAWA, S. 1935. Studies on the Soils of Manchoukuo. I. Bul. Sci. Fak. Terkult., Kjusu Imp. Univ., VI, 3.
- HIRAI, K. 1935. Studies on the Soils of Manchoukuo. II. *ibid.*
- KABURAKI, T. and MISAHA, K. 1936. Botany and Zoology in Nikko. Tôshôgû.
- KOBAYASHI, S. 1936. Earthworms from Kôryô, Korea. Sci. Rep. Tôhoku Imp. Univ., Biol., XI, 1.

- KOBAYASHI, S. 1938. Earthworms of Korea. I. *ibid.*, XIII, 2.
 — 1938 a. Earthworms from Hakodate, Hokkaidō. *Ann. Zool. Japan*, XVII, 3-4.
 MICHAELSEN, W. 1900. *Oligochaeta*. Das Tierreich. Berlin.
 — 1901. Oligochaeten der Zoologischen Museen zu St. Pétersburg und Kiew. *Bull. Acad. Imp. Sci. St.-Pétersburg*, XV, 2.
 — 1902. Neue Oligochaeten und neue Fundorte altbekannter. *Mitt. naturhist. Mus. Hamburg*, XIX.
 — 1903. Die geographische Verbreitung der Oligochaeten. Berlin.
 — 1909. The Oligochaeta of India, Nepal, Ceylon, Burma, and the Andaman Islands. *Mem. Ind. Mus.*, I.
 — 1910. Die Oligochätenfauna der vorderindisch-ceylonischen Region. *Abt. Ver. Hamburg*, XIX.
 — 1910 a. Zur Kenntnis der Lumbriciden und ihrer Verbreitung. *Ann. Mus. Acad. Imp. Sci. Pétersburg*, XV.
 — 1928. *Miscellanea oligochaetologica*. *Ark. Zool.*, 20 A.
 — 1929. Oligochäten der Kamtschatka-Expedition 1908-1909. *Ann. Mus. Acad. Sci. URSS*, XXX.
 — 1931. The Oligochaeta of China. *Peking Nat. Hist. Bull.*, V, 2.
 — 1932. Variations- und Mutationsverhältnisse bei den Arten der Lumbricidengattung *Eisenella*. *Jen. Zeitschr. Naturwiss.*, LXVII.
 — 1934. Oligochaeta from Sarawak. *Quart. Jour. Micr. Sci.*, LXXVII, 1.
 NOMURA, S. and KOBAYASHI, S. 1936. The Hydrogen Ion Concentration of the Soil of the Habitat of Earthworms. *Zool. Magaz.*, XLVIII, 8-10.
 ÔGA, I. 1931. Vegetation and Distribution of Plant in Manchoukuo. *Biol. Ser.*, Iwanami, Tôkyô.
 OHFUCHI, S. 1938. New Species of Earthworms from North-eastern Honshû Japan. *Saitô Ho-on Kai Mus. Res. Bull.*, XV.
 ÔSIN, M. 1934. Systematics and Distribution of the Family Lumbricidae in Japan. *Zool. Magaz.*, XLVI, 545.
 PICKFORD, G. E. 1926. The Oligochaeta of Wicken Fen, I. Lumbricidae. *Nat. Hist. Wicken Fen*, III.
 — 1937. A Monograph of the Acanthodriline Earthworms of South Africa. Yale Univ. press.
 SASAKI, K. 1924. *Allolobophora foetida* (Sav.) in North Japan. *Sci. Rep. Tôhoku Imp. Univ.*, Biol., I, 1.
 SCIACCHITANO, I. 1932. Fauna del Parco Nazionale del Gran Paradiso. *Parco Naz. Gran Paradiso*, III.
 — 1934. Sulla distribuzione geografica degli oligocheti in Italia. *Arch. Zool. Italiano*, XX.
 SMITH, F. 1917. North American Earthworms of the Family Lumbricidae in the Collections of the United States National Museum. *Proc. U. S. Nat. Mus.*, LII.
 — 1924. The Calciferous Glands of Lumbricidae and Diplocardia. *Illinois Biol. Monogr.*, IX.
 STEPHENSON, J. 1922. Some Earthworms from Kashmir, Bombay, and Other Parts of India. *Rec. Ind. Mus.*, XXIV, 4.
 — 1923. Oligochaeta. Fauna of British India. London.
 — 1930. Oligochaeta. Oxford.
 SVETLOV, P. 1924. Beobachtung über Oligochaeten der Regenwürmer. I. Zur Systematik.

- Fauna und Ökologie der Regenwürmer. Bull. Inst. Rech. Biol. Perm, II, 8.
- TAKAHASHI, M. 1936. An Ecological Study of Vegetation in the Province of Jehol, Manchoukuo. Rep. 1st Sci. Exp. to Manchoukuo, IV, 3.
- TÉTRY, MILE A. 1937. Révision des Lombriciens de la collection de SAVIGNY. Bull. Mus. Hist. Nat., IX.
- 1938. Révision des Lombriciens de la collection de SAVIGNY (2^e Note) *ibid.*, X.
- UDE, H. 1929. Oligochaeta. Die Tierwelt Deutschlands.
- YAMAGUCHI, H. 1930. Preliminary Note on some Earthworms from Sapporo. Zool. Magaz., XLII, 469.

MEIOTISCHE TEILUNG VON *Dictyosiphon* *FOENICULACEUS*¹

VON

KÔGORÔ ABE

*Biologische Station zu Asamushi der Kaiserlichen Tôhoku Universität,
Sendai, Japan*

Mit Tafel IX und 2 Textfiguren)

(Eingegangen am 20. März 1940)

Über die Entwicklungsgeschichte von *Dictyosiphon foeniculaceus* (HUDS.), GREV., eine Species der Dictyosiphonaceen, machte SAUVAGEAU 1917 eine interessante Mitteilung. Nach ihm keimten die in unilokulären Sporangien gebildeten Schwärmer ohne Kopulation und entwickelten sich zu Prothallien mit plurilokulären Sporangien. Aus diesen plurilokulären Sporangien traten Schwärmer hervor. Die Zygotenkeimung und die Weiterentwicklung bis zu normalen, polysiphonen Pflänzchen wurde auch von ihm ausführlich verfolgt. Durch diese Untersuchung veranlasst, schlugen SETCHELL und GARDNER (1925) die Errichtung einer neuen Ordnung Dictyosiphonales vor.

Bis jetzt liegt aber keine zytologische Untersuchung über diese interessante Alge vor. So werde ich unten meine Ergebnisse über die meiotischen Teilungen in unilokulären Sporangien von dieser Pflanze berichten.

Das Material sammelte ich im letzten Frühling während meines etwa zehntägigen Aufenthalts im Muroran Institute für Phykologie der Kaiserlichen Hokkaidô Universität. Diese Pflanze wächst immer in dieser Gegend auf *Scytosiphon lomentarius*, wie Textfig. 1 zeigt.

Das Material wurde mit der Lösung, welche ich bei der Untersuchung von *Sargassum* (ABE 1933), *Heterochordaria* (ABE 1936), *Desmarestia* (ABE 1938) und *Laminaria* (ABE 1939) verwandt hatte, meistens 5–6 Stunden lang fixiert. Die 4–5 μ dick geschnittenen Paraffinschnitte wurden mit HEIDENHAIN'S Eisenalaunhämatoxylin gefärbt.

Die Fortpflanzungsorgane gehen aus einer Oberflächenzelle des Thallus hervor. Fig. 1 zeigt den sich im vollständigen Ruhestadium befindlichen Kern einer solchen Zelle. In diesem Stadium beträgt der Durchmesser des Kerns etwa 3 μ . In Figg. 3–4 sieht man ein Synapsisstadium. Darauf folgt Spirem- und Diakinesestadium (Figg. 5–6). In Fig. 6 ist

¹Contribution from the Marine Biological Station, Asamushi, Aomori ken No. 167.

der Nukleolus schon verschwunden. Bei solchen Stadien konnte ich feststellen, dass die Zahl der Chromosomen mit grosser Wahrscheinlichkeit 18 beträgt. Vor der Auflösung der Kernmembran ordnen sich die Chromosomen allmählich auf der Äquatorialebene an (Fig. 7). Die vollständige Metaphase erfolgt aber nach Auflösung der Kernmembran. Fig. 8 gibt dasselbe Stadium in Polansicht wieder, wobei sich 18 Chromosomen klar zählen lassen. In der Seitenansicht der Spindel ist kein

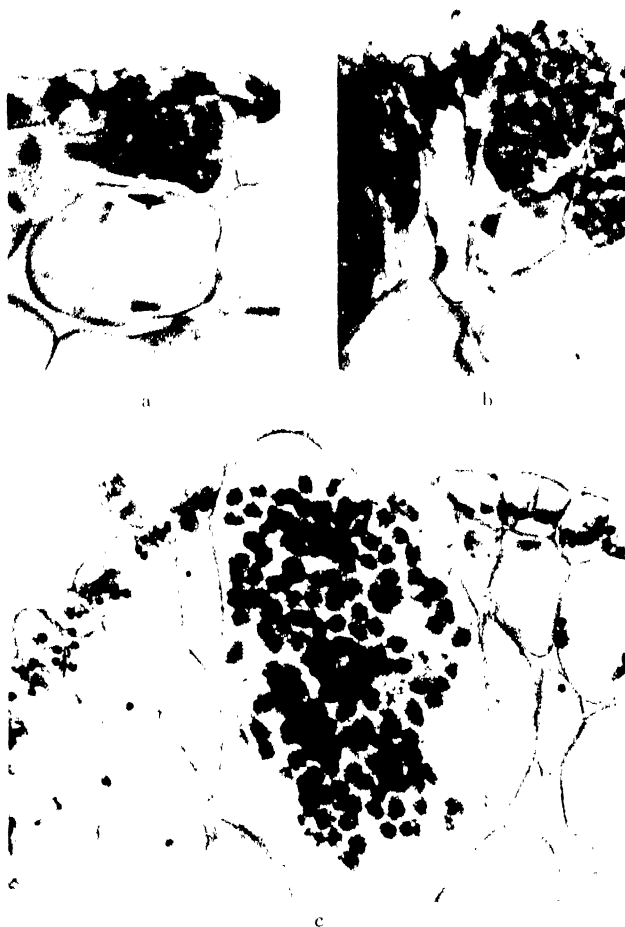


Textfig. 1. *Dictyosiphon foeniculaceus* auf *Scytosiphon lomentarius*. Verkl. 1/4.

zentrosomähnliches Körperchen in jedem Pole sichtbar (Fig. 9). Die Anasowie Telophase geht normal vor sich (Figg. 10–11). Nach dieser Teilung entstehen natürlich vier Kerne, die dann noch weitere simultane Teilungen ausführen. Inzwischen vergrössert sich das Sporangium allmählich (Textfigg. 2, a b). Und am Ende teilt sich das Protoplasma auf; jede Portion enthält einen Kern und einen Chromatophor und entwickelt sich später zu Schwärmern (Textfigg. 2, c).

Aus obigen Resultaten ist es sicher, dass die Individuen mit unilokulären Sporangien diploid und die zwei ersten Kernteilungen im Sporangium Reduktionsteilungen sind. *Dictyosiphon foeniculaceus* zeigt also einen regel-

mässigen Generationswechsel zwischen einer plurilokuläre Sporangien tragenden zwerghaften haploiden und einer unilokuläre Sporangien tragenden stattlichen diploiden Generation.



Textfig. 2. Unilokuläres Sporangium. *a-b* vielkernige Stadien, *c* voll reifes Stadium. Vergr. 560.

Es sei mir an dieser Stelle erlaubt, meinem hochverehrten Lehrer, Herrn Prof. Dr. M. TAHARA, meine grosse Dankbarkeit auszusprechen für die Anregung zu dieser Arbeit. Ebenso bin ich auch Herrn Prof. Dr. Y. YAMADA und Herrn T. KANDA für ihre liebenswürdige Unterstützung beim Sammeln des Untersuchungsmaterials zu grossem Dank verpflichtet.

LITERATURVERZEICHNIS

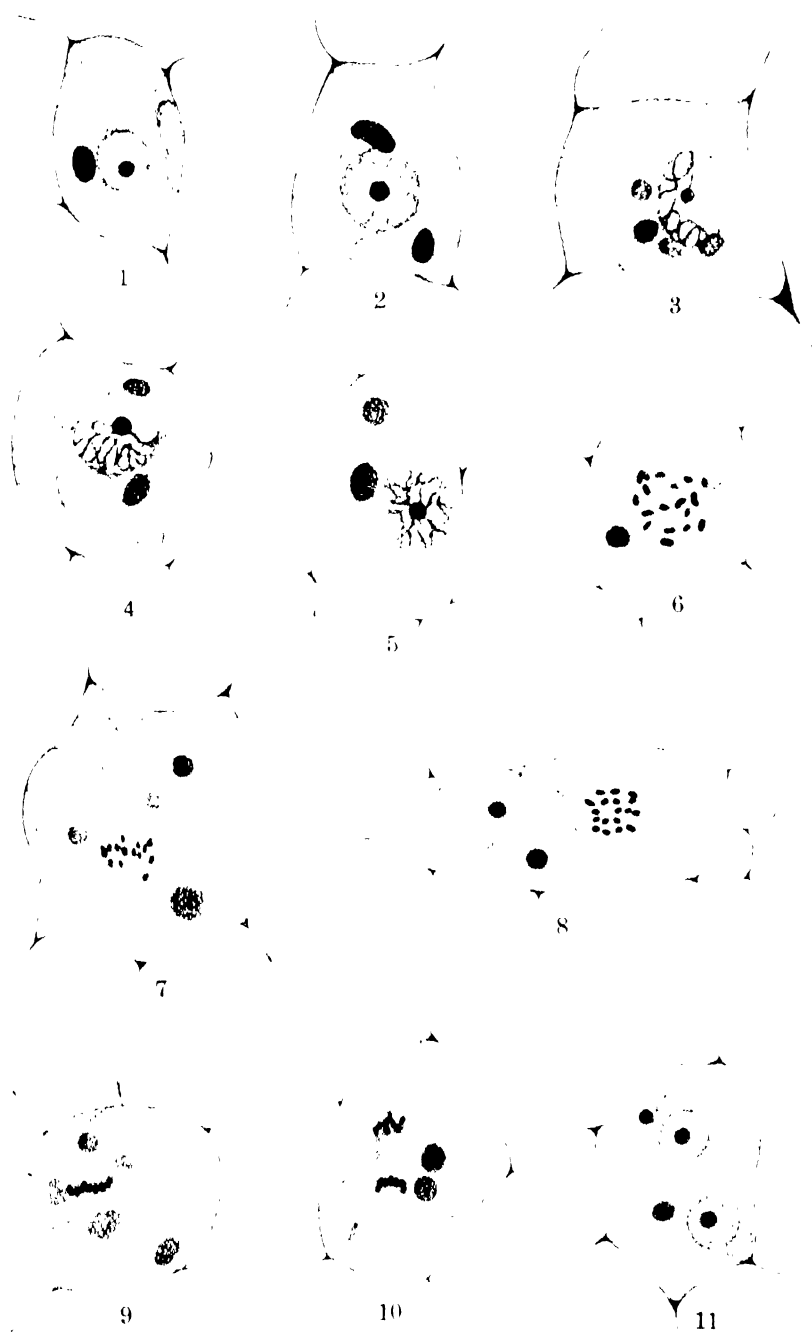
1. ABE, K., 1933. Mitosen im Antheridium von *Sargassum confusum*. Sci. Rep. Tôhoku Imp. Univ., Biol., Vol. VIII.
2. ABE, K., 1936. Kernphasenwechsel von *Heterochordaria abietina*. Sci. Rep. Tôhoku Imp. Univ., Biol., Vol. XI.
3. ABE, K., 1938. Entwicklung der Fortpflanzungsorgane und Keimungsgeschichte von *Desmarestia viridis* (MÜLL. LAMOUR. Sci. Rep. Tôhoku Imp. Univ., Biol., Vol. XII.
4. ABE, K., 1939. Mitosen im Sporangium von *Laminaria japonica* ARESCH. Sci. Rep. Tôhoku Imp. Univ., Biol., Vol. XIV.
5. SAUVAGEAU, C., 1917. Sur un nouveau type d'alternance des générations chez les algues brunes *Dictyosiphon foeniculaceus*. Comt. rend. de l'Acad. de Sci., Tome 161.
6. SAUVAGEAU, C., 1929. Sur le développement de quelques Phéosporées. Bulletin de la Station Biologique d'Arcachon, Tome 26.
7. SETCHELL, W. A. and GARDNER, N. L., 1925. The marine algae of the Pacific Coast of North America. III. Melanophyceae. Univ. of Calif. Publ. Bot., Vol. 8.

TAFELERKLÄRUNG

Alle Figuren wurden mit Hilfe eines Abbéschen Zeichenapparatus gezeichnet unter Benutzung des ZEISSchen Objektiv, Ölinmersion 1/12 und des ZEISSchen Okular $\times 17$. Vergr. $\times 2600$.

TAFEL IX

Fig. 1. Ruhestadium. Fig. 2. Früheste Prophasestadium. Figg. 3-4. Synapsisstadium. Fig. 5. Spiremstadium. Fig. 6. Diakinese. Fig. 7. Frühere Metaphase in Seitenansicht. Fig. 8. Vollständige Metaphase in Polansicht. Fig. 9. Vollständige Metaphase in Seitenansicht. Fig. 10. Anaphase. Fig. 11. Telophase.



ON THE DEVELOPMENT OF THE CONCEPTACLE OF *SARGASSUM*, *COCCOPHORA* AND *CYSTOPHYLLUM*

By

MASATO TAHARA

Biological Institute, Tôhoku Imperial University, Sendai, Japan

(With 7 Text figures)

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The conceptacle of the Fucaceae is one of the most important characteristics of these plants. Our present knowledge concerning the development of this organ is founded on SIMONS' researches with regard to *Sargassum filipendula* in the year 1906. More recently it has been proved by NIENBURG (1913) that the mode of the development of the conceptacle varies somewhat in different members of the family. He was restricted, however, in his choice of materials, which were mainly the plants easily accessible to him. So that there remains a number of important genera which need to be investigated in this respect. *Sargassum*, *Cystophyllum* and *Coccophora* are very common Fucaceous algae found along the coast of Japan. So the present writer has followed the development of the conceptacle of these plants in materials newly obtained in the spring of this year. It was rather difficult to know exactly when these plants attain the developmental stages at which it was so necessary to examine them. The writer visited the Misaki Marine Biological Station several times in order to collect the materials. It is his pleasant duty to express here his gratitude to Prof. Y. OKADA and Prof. E. ERI for the facilities given him during his stay in that institution.

Coccophora is an alga growing only along the coast of the Japan Sea. The writer wishes to acknowledge his indebtedness to Dr. K. ABE, who kindly collected the material of this plant at the Asamushi Marine Biological Station.

The materials were fixed with chrom-acetic solution of different constitutions, with or without osmic acid. For staining of the cell wall ruthenium red was very effective.

1. *Sargassum enerve* Ag.

Sargassum enerve is one of the most common species of *Sargassum* found along the coast of Japan. The materials obtained at Misaki from the middle of January to the end of February were very useful.

The initial cell of the conceptacle lies near the apical cell of a receptacle.

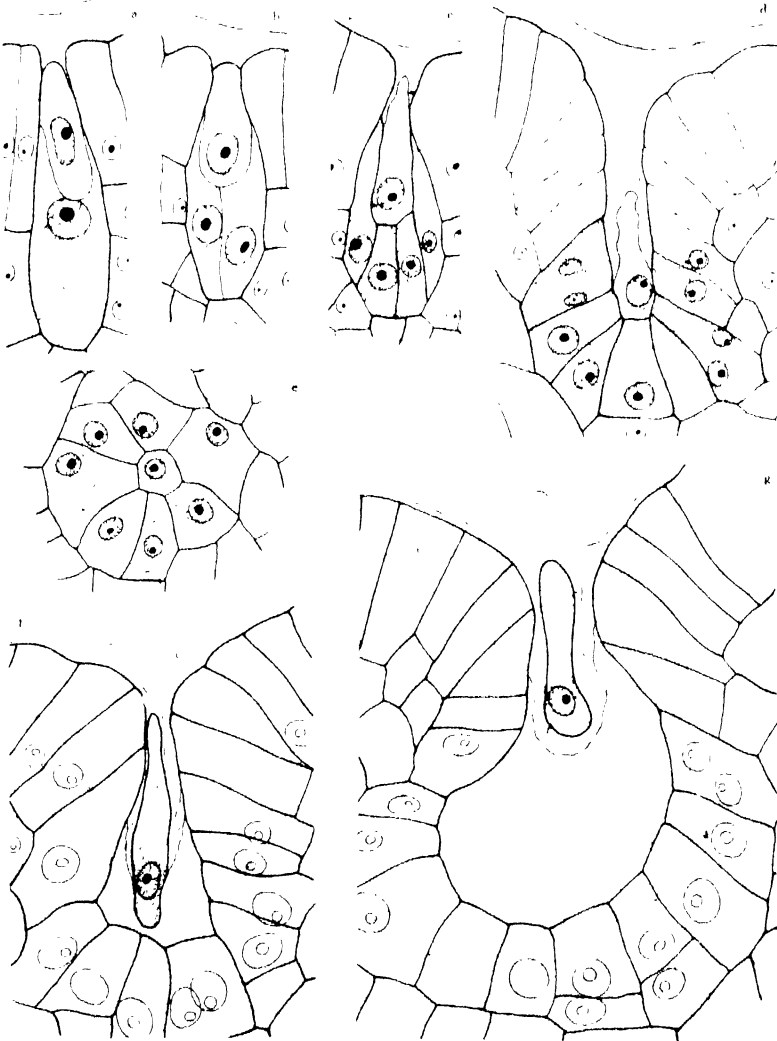


Fig. 1. Conceptacle development in *Sargassum enerve*. *a*, initial cell in two cell stage. *b d*, longitudinal divisions of the lower cell. *e*, cross section through the basal part of a conceptacle. *f--g*, dislocation of the tongue/cell. $\times 900$.

As is already known, the development of a conceptacle is initiated by the activity of this cell. In the first place the cell is divided transversely into two cells by a curved wall with a concave surface above. Following SIMONS' nomenclature the upper cell of this stage is called the tongue cell. In the present species, in the whole course of the development of the conceptacle, no division occurs in this cell. The lower cell is, however, the more important cell in the development of the conceptacle. It is divided longitudinally into two similar cells by a vertical wall. The third division-wall is also vertical and is perpendicular to the second.

In the succeeding stages the longitudinal divisions continue, until the principal portion of the flask-shaped conceptacle is formed by the products of this lower cell. It is significant that in the basal part of the conceptacle the wall cells are arranged radially around the centrally situated tongue cell.

So far the process agrees in substance with SIMONS' descriptions concerning the development of the conceptacle of *Sargassum filipendula*. She does not give, however, further details of the tongue cell. According to the writer's observation, in *Sargassum enerve* this cell which is found, for a time after its formation, fixed to the basal part of the conceptacle, later becomes free from the general wall of the conceptacle and is transferred towards the mouth of the conceptacle. A mass of gelatinous substance is secreted around the tongue cell, completely closing up the mouth of the conceptacle. In a conceptacle which has nearly completed the whole course of its development, the tongue cell can no more be distinguished with certainty. In this species the rather important part of the conceptacle is formed by the aggressive growth of the general epidermal tissue. In this respect this species differs from *Sargassum filipendula*.

2. *Sargassum Horneri* Ag.

This alga is also very common along the coast of Japan. The initial cell of the conceptacle divides also at first into two dissimilar cells by a curved wall. The upper cell of the two, the tongue cell, remains undivided. The lower cell divides repeatedly by longitudinal walls. The arrangement of the wall cells around the tongue cell is also radial.

So far the mode of the development is about the same as seen in the foregoing species. But in the present species, the tongue cell enlarges, keeping pace with the growth of the conceptacle and fills up the inner cavity of the conceptacle. So, at first no space is seen between the tongue

cell and the inner wall of the conceptacle. Meanwhile the plasmic content of the cell is thrust into the upper portion of the cell. And a new wall is formed at the base of the tongue cell, thus giving birth to the spacious

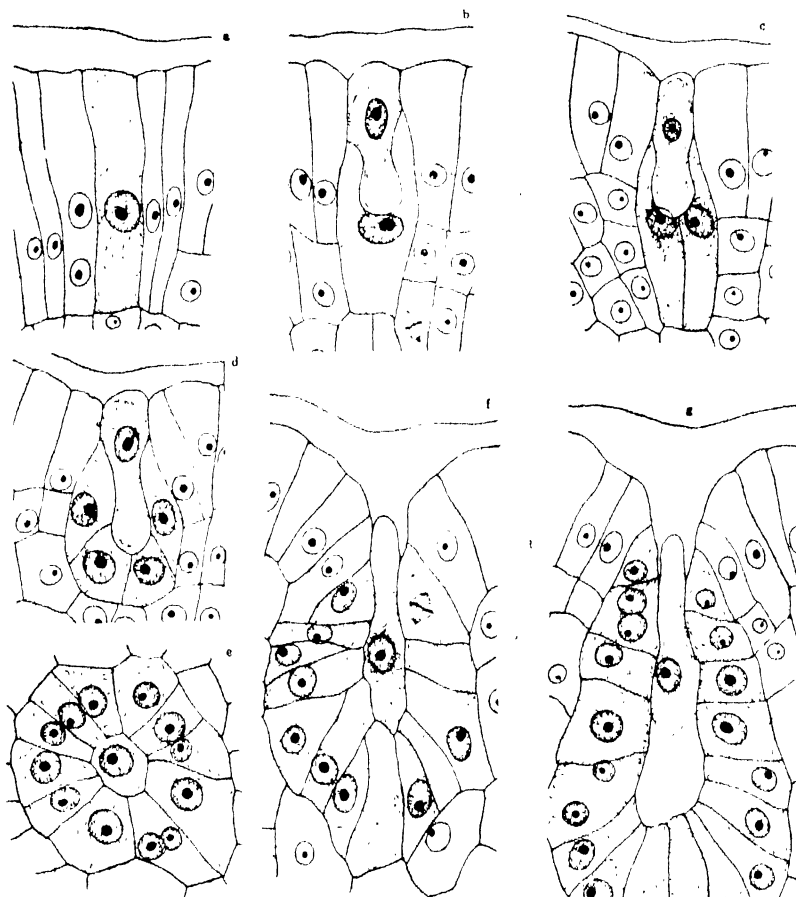


Fig. 2. Conceptacle development in *Sargassum Horneri*. *a*, initial cell. *b*, two cell stage of the same. *c*—*d*, divisions of the lower cell. *e*, cross section through the basal part of a conceptacle. *f*—*g*, further stages of development: tongue cell grows, filling the inner cavity of the conceptacle. $\times 900$.

cavity of the conceptacle. By the growth of the inner wall of the conceptacle, the cavity enlarges gradually. The tongue cell remains intact in these stages and takes the appearance of the stopper of the cavity of the conceptacle.

The secretion of gelatinous substance around the tongue cell is not so

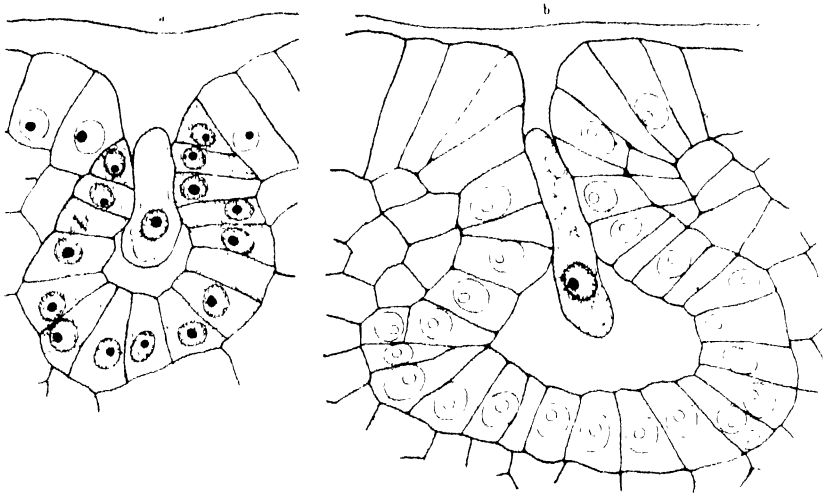


Fig. 3. Conceptacle development in *Sargassum Horneri*. Tongue cell becomes free from the wall of the conceptacle. $\times 900$.

conspicuous as in the preceding species. In the final stage of the development the tongue cell cannot be distinguished with certainty. As is clearly seen from the figures, the inclination of the earlier wall-cells of this plant is rather significant compared to that of the foregoing species. In the present species, as in *Sargassum filipendula*, almost the whole inner wall of the conceptacle is formed by the products of the lower cell of the two cell stage.

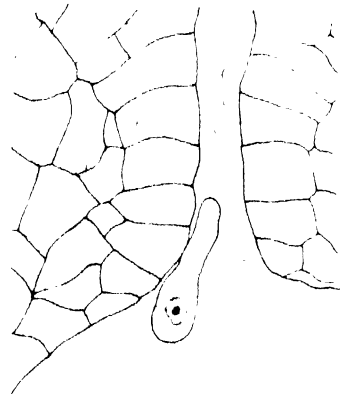


Fig. 4. *Sargassum Horneri*. Tongue cell is beginning to degenerate. $\times 900$.

3. *Coccophora Langsdorfii* GREV.

When the reproductive organs of this plant have attained maturity the vesicles are already absent. And hollow receptacles take the role of the vesicles. But in the first stage of the development the receptacle is still solid. Such receptacles showing the youngest stage of the conceptacle development are much larger than those of *Sargassum* in the same stage of development.

The process of the conceptacle development is at first just the same,

as seen in *Sargassum*. The arrangement of the wall cells around the tongue cell is also radial. But in the following stages of development things are peculiar to this plant.

1. In the course of development the tongue cell divides transversely and takes the form of a typical paraphysis.

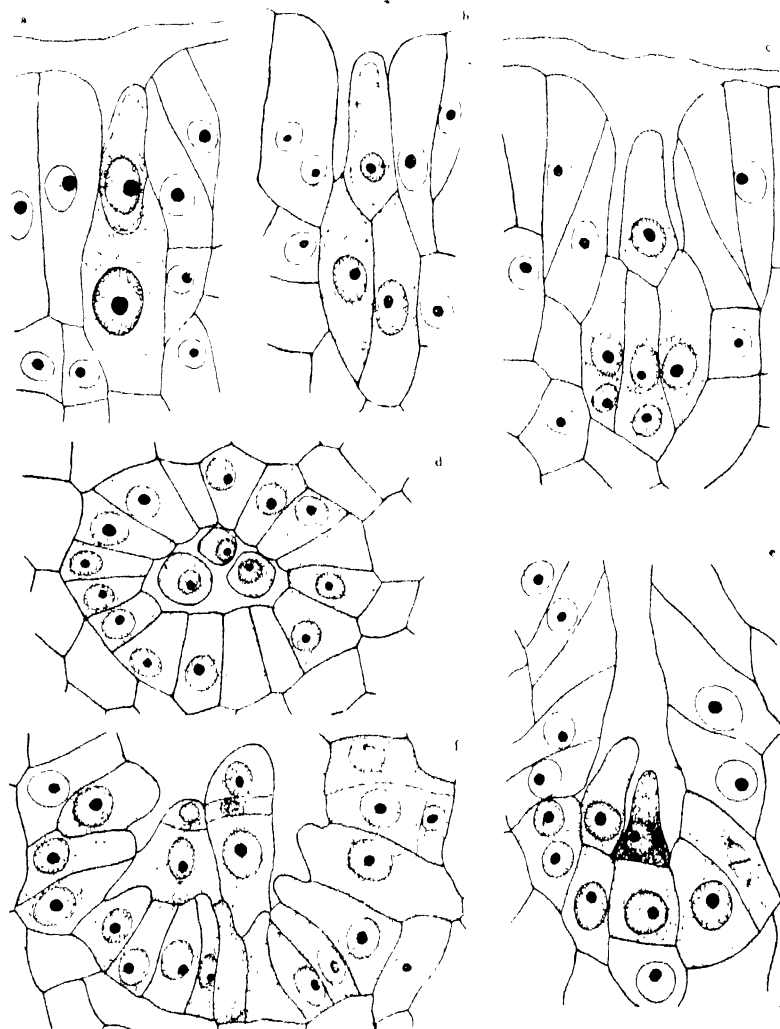


Fig. 5. Conceptacle development in *Coccophora Langsdorfi*. *a*, initial cell in two-cell stage. *b*–*c*, divisions of the lower cell. *d*, cross section through the basal portion of a conceptacle. *e*–*f*, transverse divisions of the tongue cell and growth of a wall-cell into the cavity of the conceptacle. $\times 900$.

2. In an early stage of conceptacle development, a few of the wall-cells begin to elongate towards the central cavity of the conceptacle and form paraphyses by transverse division, in a manner similar to that of the tongue cell. So in a cross section of the basal part of a young conceptacle, we often see 2 to 4 similar cells at the center of the conceptacle cavity.

Transverse divisions of the tongue cell were occasionally observed by SIMONS in the cryptostomata of *Sargassum filipendula*. And its regular occurrence is reported by NIENBURG in the conceptacles of *Cystosira* and *Halidrys*.

In a mature conceptacle of *Coccophora*, we see multitude of slender paraphyses. These are, however, totally different in shape and structure from those primary paraphyses developed in an early stage of development from the tongue cell and wall-cells. Each of the latter consists of a series of flattened cells, thus having an appearance like a bamboo shoot (Fig. 6).

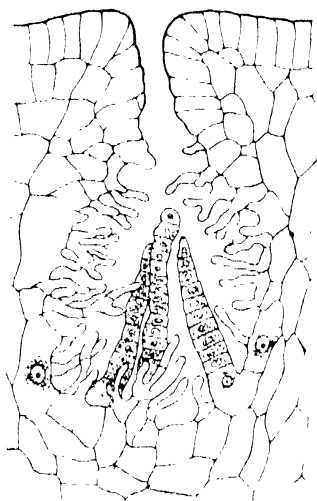


Fig. 6. Conceptacle of *Coccophora Langsdorffii* in a later stage of development. $\times 200$.

4. *Cystophyllum sisymbrioides* AG.

In Misaki, this alga is also common as the former two species of *Sargassum*. These three species ripen in about the same season. The early stages of the conceptacle development is about the same, as seen in *Sargassum* and *Coccophora*. The initial cell of the conceptacle is divided into two dissimilar cells by a curved, transverse wall, the upper cell forming the tongue cell. The walls which are formed in successive longitudinal divisions of the lower cell are rather vertical. Fig. 7, *d* shows a cross section of the basal part of a young conceptacle. It is remarkable that in this case the cells resulting from the longitudinal divisions of the lower cell are not arranged radially around the central tongue cell; the cell-walls cross each other mostly at right angles. As has been already described by the present writer in a previous paper (1913), the conceptacle in the female of this plant has in the time of the oogonium liberation no opening towards the outside. The closure of the conceptacle is carried

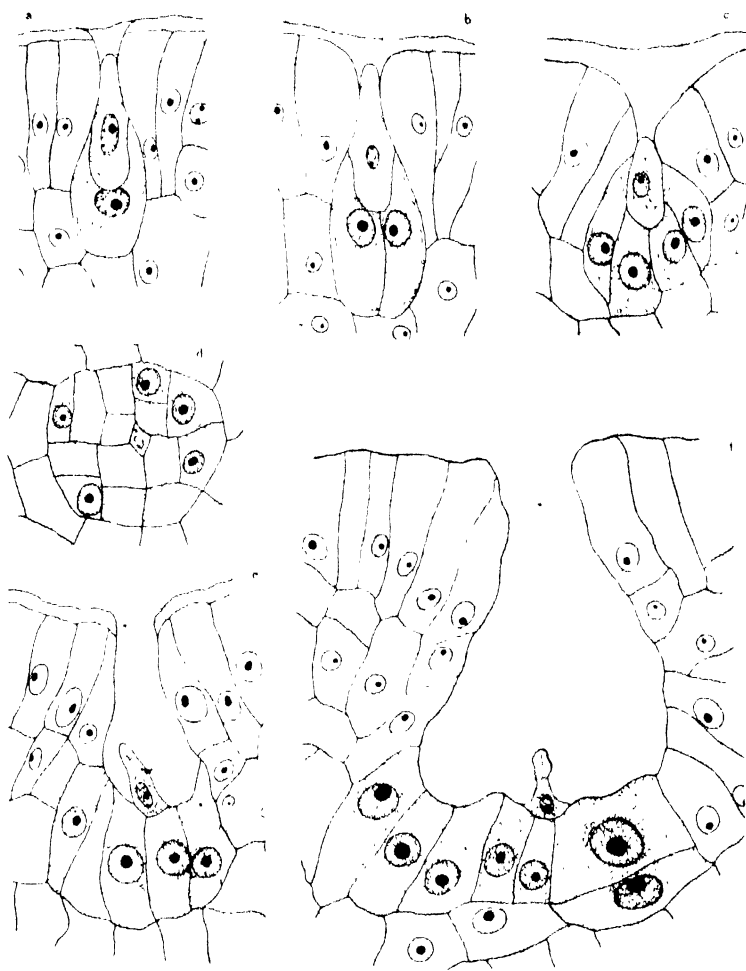


Fig. 7. Conceptacle development in *Cystophyllum sisymbrioides*. a, initial cell in two cell stage. b—f, further stages of development. d, cross section through the base of a conceptacle. $\times 900$.

out by the growth of the epidermal tissue in a later stage of the conceptacle development.

DISCUSSION

In the present research the writer has investigated the conceptacle development in two species of *Sargassum* and one species of *Coccophora* and *Cystophyllum*. Whether the dislocation of the tongue cell, which was

observed in the two species of *Sargassum* is a general characteristic or no of the genus *Sargassum* will be decided by a future investigation. But it is surprising that this significant phenomenon is not mentioned by SIMONS in *Sargassum filipendula*.

In any case it is remarkable that the closing up of the passage of the conceptacle by the aid of the tongue cell is found only in *Sargassum*, the most highly differentiated genus in the Fucaceae. The temporary closure of the young conceptacle of both sexes may be looked upon as a provision of nature essential for the protection of the delicate meristematic tissue of the conceptacle.

In the paper above cited, NIENBURG has pointed out that the inclination of the longitudinal walls of the cells around the central tongue cell is different in *Sargassum*, *Cystosia*, *Halidrys* and *Pycnophycus*. But as is shown in the present investigation the inclination of the wall-cells is different in two species of *Sargassum*. According to the writer's opinion, the arrangement of the wall-cells around the tongue cell, which is clearly seen in a cross section through the basal part of a young conceptacle, appears to have a more important significance. It is radial in *Sargassum* and *Coccophora* and rectangular in *Cystophyllum*.

SUMMARY

1. In *Sargassum enerve* the initial cell of the conceptacle divides at first by a curved wall. The upper cell, i.e. the tongue cell remains undivided through the whole course of development. In the lower cell longitudinal division is repeated and it forms the principal part of the conceptacle. The tongue cell remains for a time fixed to the wall of the conceptacle, but later it becomes free and is transferred towards the mouth of the conceptacle. In this stage, the secretion of gelatinous substance around the tongue cell is very conspicuous. Thus the passage of the conceptacle is completely closed up. In a conceptacle which has nearly completed its development, the tongue cell can no more be distinguished.

2. The conceptacle development in *Sargassum Horneri* is in substance similar to that in *Sargassum enerve*. But one point is worth mention. The tongue cell of this plant grows keeping pace with the growth of the conceptacle and filling the cavity of the conceptacle. So, for a time there is no space between the tongue cell and the inner wall of the conceptacle. But meanwhile after becoming free from the wall of the conceptacle the tongue cell is transferred to the mouth of the conceptacle and has the function of the stopper of the conceptacle.

3. In *Coccophora Langsdorfii* the tongue cell divides transversely and forms a paraphysis. It is remarkable that in this species a few of the wall-cells of the conceptacle grow, in an early stage of the conceptacle development, towards the cavity of the conceptacle and become paraphyses, dividing transversely just like the tongue cell.

4. A cross section through the basal part of a young conceptacle shows clearly the arrangement of the wall-cells around the centrally situated tongue cell. This is radial in *Sargassum* and *Coccophora*, and is not radial in *Cystophyllum*, in this case the walls being formed as to cross each other at right angles.

LITERATURE

- BOWER, F. O. (1880). On the Development of the Conceptacle in the Fucaceae. *Quar. Jour. Micr. Sci.* 20.
- DELF. MARION (1939). The Systematic Position of the Fucales. *New Phyt.* 38.
- NIENBURG, W. (1913). Die Konzeptakelentwicklung bei den Fucaceen. *Zeit. Bot.* 5.
- SIMONS, E. B. (1906). A Morphological Study of *Sargassum filipendula*. *Bot. Gaz.* 41.
- TAHARA, M. (1913). Oogonium Liberation and the Embryogeny of some Fucaceous Algae. *Jour. Coll. Sci., Imp. Univ. Tokyo.* 32.

LIMNOLOGICAL STUDY OF LAKE OSORESAN-KO A REMARKABLE ACIDOTROPHIC LAKE IN JAPAN*

By

KIKUYA MASIKO

The Marine Biological Station, Asamushi, Japan

(With 10 Text-figures)

(Received May 13, 1940)

I. INTRODUCTION

Lake Osoresan-ko, or Usori-ko, lies in the northernmost district of Honshû, filling up the caldera of the Volcano Osore-san, Simokita Peninsula, Aomori Prefecture.

Lake Osoresan-ko as far as it has been studied is one of the most remarkable lakes in Japan. Despite the high acidity of its water the yields of this lake are fairly rich, though the number of different species of life met with in it is relatively small. From the summer of 1931 until 1934 a number of investigations of this lake were carried out by several scientists. The writer also has made some observations of the lake during the period extending from 1935 to 1938. In the present paper the writer wishes to deal with the results thus far obtained and also to compare them with those obtained by other scientists.

The lake lies at an altitude of about 300 meters. The area covered by the lake water is 2.17 square km (according to a map of 1/200,000 scale, of the Land Survey Department). The lake empties its water into the Pacific Ocean through the only outlet, River Syôzu-gawa (or Sanzuno-kawa) in the north; and is fed by several small rivers coming from the surrounding mountains. There are also many small streams flowing into it which have their source in hot-springs found to north of the lake, and these constantly supply the strong acidic water. Moreover there are many small hot-springs found on the lake-beach itself as well as in the shallow bottom of the lake and the water close to these hot-springs is often highly acidified and contains little or no oxygen.

* Contribution from the Marine Biological Station, Asamushi, Aomori-Ken, No. 168.

II. PHYSICAL AND CHEMICAL CONDITIONS OF THE LAKE WATER

The general features of the physical and chemical conditions of this lake were observed during the time extending from 1931 to 1934 by several writers, and are shown in Table I. The results obtained by the present writer during the period from 1936 to 1938 are shown in Table II and III, being included also the data secured by TEZUKA.

a) Transparency and Colour of Water

Some records to show the transparency and colour of the water in the lake were obtained and they are tabulated below :

TABLE I.

Date	Transp. (m)	Colour	Date	Transp. (m)	Colour
1931, 24/IX	5	15.0	1935, 15/IX	5.5	8.5
1932, 14/V	9	1.5	16/IX	6	7.5
22/VI	3.5	14.0	1936, 4/VIII	8	5.0
13/VIII	3.5	14.0	1937, 3/VII	—	4.0
24/XI	10	2.5	1938, 28/VI	6	5.5
1934, 30/VIII	6	4.2	13/VIII	8	5.0
29/IX	5	8.0			

From the above table we learn that the water has not shown any remarkable decrease of transparency since 1934. TAMURA recorded such high transparency as 15 and 14 meters in 1931 and 1932 respectively, while the highest value obtained during the period from 1934 to 1938 was 8.5 meters as observed on September 15, 1935. The colour of the water, which was measured according to FOREL's scale, was somewhat lower than in former years. The striking low value obtained in May of 1932 may be considered as being due to contamination from thawing water, as was stated by TAMURA (1933). Concerning the low value shown in November, TAMURA thought that it was due to the increase of the inflow of water which took place in the autumn as in the case of Lake Tazawako mentioned by YOSHIMURA in 1932.

The colour of the water seemed usually to vary from No. 6 to 8 in the years since 1934, while it was No. 3.5 in the summer of 1932. Though the colour is shown according to FOREL's scale in the above, in reality it appears somewhat whitish. This is also the fact in other anorganic aciditrophic lakes, such as Lake Kata-numa, L. Itibisinai-ko, and three lakelets of Bandai, Ruri-numa, Ao-numa, and Benten-numa. With regard

to this fact, YOSHIMURA stated that it may depend upon the diffused light caused by the colloidal suspension of some substance of the nature of calcium sulphate.

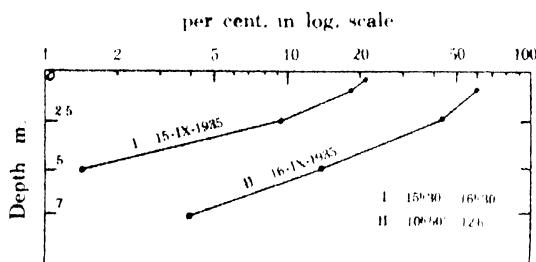


Fig. 1. Measurement of submarine illumination.
Ordinate....Depth in meter.
Abcissa....Percentage illumination.

The data shown in the Fig. 1 are the results of a measurement obtained by means of hydroiodic acid to ascertain the light intensity penetrating into the lake-water. The measurement was made after PEARSALL's method, but the writer considers that MCCREA's method may be more suitable for general cases (PEARSALL, 1921; MCCREA, 1923).

b) Water Temperature

Most of the investigations hitherto made have been carried out during summer time. With regard to the temperature of surface water no remarkable differences were recognized every year. The highest temperature of the surface water was observed on Aug. 13, 1938 (Fig. 5), and the next highest on Aug. 13, 1932 (Fig. 2), it was respectively 24.7° and 24.0°. On Aug. 12, 1931 the surface temperature was 22.2° and on Aug. 1, 1936 (Fig. 4), it was 22.6°. On the other hand, the temperature of deeper water has shown much difference every year, as seen in Table II and III. The most noticeable fact is that in the summer of 1931 and 1932 stratification in the lake-water was scarcely recognized, while in the summers of from 1934 to 1938 very distinct stratification was observed. YOSHIMURA pointed out that TAMURA could not find any stratification in the lake water during the years 1931 and 1932 because he had omitted to test the deepest basin (1934 a). Comparing the data obtained by YOSHIMURA in the year 1934 with those of the proceeding years obtained by TAMURA, however, it is obvious that the lake-water showed a distinct stratification in the summer of 1934. In the summer of 1931 no thermocline was

found from the surface to the bottom (11 m deep) and in 1932 the case was almost the same, while on Aug. 30, 1931, YOSHIMURA observed a sharp thermocline occurring between the depths of 12 and 13 meters. Even on the 29th of November of that year, stratification remained in the deeper layer. And in the observation made by the writer in 1936, the summer stagnation was so distinct that the difference in the water temperature between the surface and bottom layers was 11.95° on June 28 and 11.1° on Aug. 4 respectively, and the anaerobic layer rose to within 10 meters from the surface on both days. In the summers of 1937 and 1938 stratification was also distinct, viz., the difference of temperature existing between the surface and bottom water was 7.7° (0-15 m) on July 4, 1937, 6.75° (0-14 m) on June 28, 1938, and 10.7° (0-13 m) on Aug. 13, 1938.

TABLE II.

1931 ⁽¹⁾ 12/VIII				24/IX			
Dep. (m)	Temp.	pH	O ₂ (cc/L)	Dep. (m)	Temp.	pH	O ₂ (cc/L)
0	22.2	3.6	6.44	0	19.8	3.3	5.44
7	19.6	3.6	6.07	6	19.0	3.3	5.50
14	19.4	3.7	6.21	12	19.0	3.3	4.46

1932 ⁽²⁾ 14/V			22/VII			13/VIII			24/XI		
Dep.	Temp.	pH	Temp.	pH	O ₂ (cc/L)	Temp.	pH	O ₂ (cc/L)	Temp.	pH	O ₂ (cc/L)
0	9.6	3.2	21.5	3.2	5.44	24.0	3.2	5.49	7.8	3.2	7.24
5	9.6	3.2	19.6	3.4	5.29	22.8	3.4	4.30	7.8	3.2	7.43
9	—	—	—	—	—	21.0	3.4	4.37	—	—	—
10	9.5	3.4	18.8	3.4	5.35	—	—	—	7.8	3.3	—
13	—	—	18.0	3.4	4.13	20.3	3.6	4.45	—	—	—
14	9.1	3.6	—	—	—	—	—	—	7.5	3.3	7.40

1934 ⁽³⁾ 30/VIII				29/IX ⁽⁴⁾			
Dep.	Temp.	pH	O ₂ (cc/L)	Dep.	Temp.	pH	O ₂ (cc/L)
0	22.0	3.0	5.61	0	17.5	3.6	5.77
5	21.7	3.05	5.90	5	17.5	3.6	5.67
10	21.5	3.15	5.50	10	17.5	3.6	5.51
14	15.0	4.8	0.00	14	15.8	4.5	1.90
15	13.3	5.0	0.00	—	—	—	—

⁽¹⁾ measured by KOKUBO et al. ⁽²⁾ measured by TAMURA, ⁽³⁾ measured by YOSHIMURA.

⁽⁴⁾ measured by TAMURA.

TABLE III.

1936 28/VI					4/VIII				
Dep.	Temp	pH	O ₂ (cc/L)	O ₂ (%)	Dep.	Temp.	pH	O ₂ (cc/L)	O ₂ (%)
0	21.2	3.8	6.86	107.8	0	22.6	3.5	5.46	88.1
2	19.1				2	22.3	3.5	5.56	89.2
4	18.5	4.4			4	22.0	3.5	5.68	90.7
5	18.2				6	21.8	3.5	5.61	89.8
6	16.5	4.6	7.35	105.1	7	19.9	3.5		
8	11.5	4.8	6.69	86.1	8	17.3	3.5	6.65	96.9
9	10.4	4.5?	4.21	54.5	9	15.2	3.5	3.30	46.5
10	10.2	3.8	0.00	0	10	13.6	3.5	0.00	0
13	9.25	4.4	0.00	0	12	12.7	3.6	0.00	0
					14	11.5	3.8	0.00	0

3/VII/1937*				28/VI/1938*			
Dep.	Temp.	O ₂ (cc/L)		Dep.	Temp.	pH	O ₂ (cc/L)
0	19.8	5.45		0	19.05	4.0	5.42
5	19.8	5.60		3	16.70	4.0	5.37
8	19.75	5.45		5	16.40	4.1	5.66
10	15.3	3.30		8	16.25	4.5	5.85
10.5	14.6	0.35		10	15.60	4.4	4.46
11	14.2	0.00		11	15.00	4.1	4.38
13	13.5	0.00		12	14.62	4.0	5.48
15	12.1	0.00		12.5	13.45	3.6	0.00
				13	13.40	3.8	0.00
				14	12.30	4.0	0.00

13/VIII/1938 15 h.					22 h.		
Dep.	Temp.	pH	O ₂ (cc/L)	O ₂ (%)	Temp.	O ₂ (cc/L)	O ₂ (%)
0	24.7	3.5	5.62	94.1	24.7	5.65	94.6
2	24.5	3.5	5.51	92.0	24.6	5.42	90.7
5	23.75	3.45	6.84	112.4	23.6	6.68	109.4
6	23.0						
8	20.4	3.45	5.28	81.9	20.5	5.78	89.7
9	18.7	3.45	0.00	0			
10	17.2	3.45	0.00	0			
11	16.3	3.55					
12	14.8						
13	14.0	3.9	0.00	0			
Syôzu-gawa 16 h.					O ₂ (%) : after Fox-Whipple-Whipple), with correction for the altitude after Palace.		
0	24.7	3.5	5.54	91.8			

* observed by TEZUKA and is mentioned in his MS.

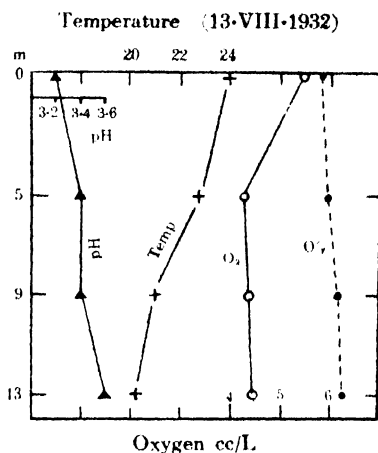


Fig. 2. Vertical distribution of temperature, O_2 and pH (Lake Osore-san-ko).

Abscissa (upper).....Temperature and pH.

Abscissa (below)....Oxygen.

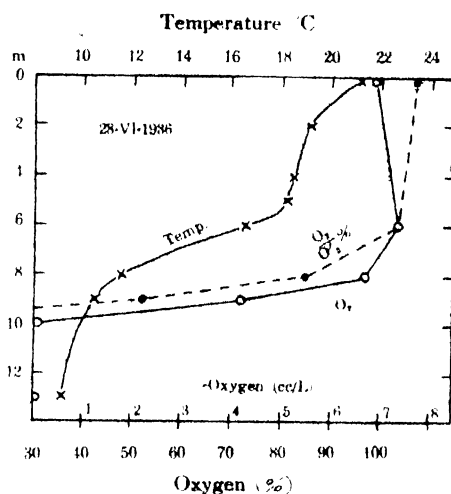


Fig. 3. Vertical distribution of temperature and Oxygen.

Abscissa (upper).....Temperature.

Abscissa (below)....Oxygen.

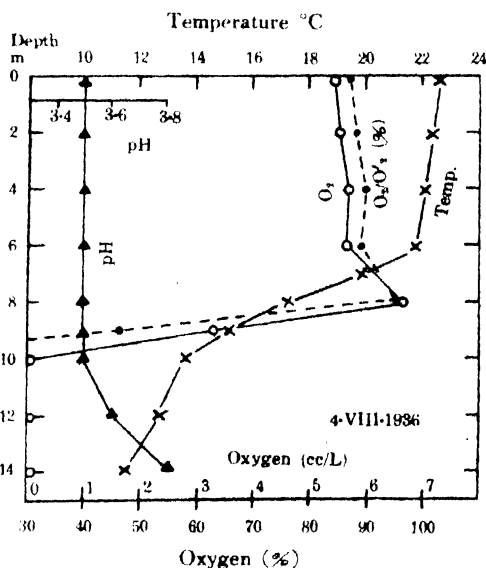


Fig. 4. Vertical distribution of temperature, O_2 and pH.

OrdinateDepth in meter

Abscissa (upper).....Temperature and pH.

Abscissa (below)....Oxygen.

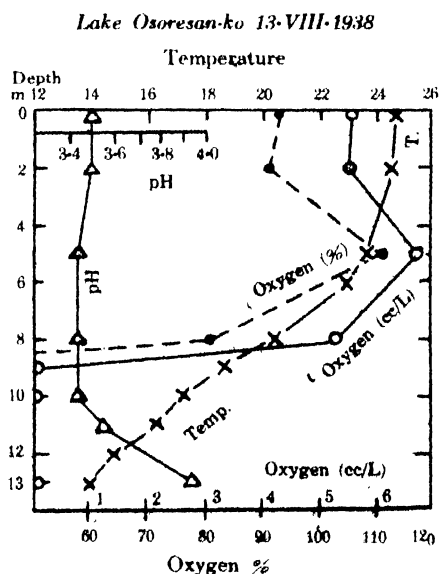


Fig. 5. Vertical distribution of dissolved oxygen.

OrdinateDepth in meter

Abscissa (upper).....Temperature and pH.

Abscissa (below)....Oxygen.

c) Hydrogen Ion Concentration

It was reported by YOSHIMURA (1934 a) that Lake Osoresan-ko is one of the most remarkable acid-water lakes in the world, and that the strong acidity is due to the inflow of sulphuric acid from the numerous hot-springs found on the northern shore of the lake. In the period of summer stagnation the pH-value is in general lowest at the surface, and it becomes rapidly higher in the lower layers. The lowest value of pH hitherto observed is 3.0 measured by YOSHIMURA (1934 a) on Aug. 30, 1934. The highest value of pH of the surface water is 4.0 being measured by TAMURA on June 28, 1938. In the case of the bottom water, a value of pH as high as 5.0 was reported by YOSHIMURA in 1934. The commonest condition of pH seems to be 3.5. The lower value of pH seen in the upper layer is obviously due to the inflow of acid water of a higher temperature derived from the hot-springs. There is, however, a noticeable phenomenon that a very irregular stratification of pH was more than once observed, viz., one on June 28, 1936, and the other on June 28, 1938. As these observations were made by students, the writer somewhat doubts the accuracy of the data; but the result obtained by TEZUKA (MS) on the same day in 1938 shows a similar anomalous condition of stratification. The writer considers that such a special condition takes place in the early stage of the summer stagnation period. Further discussion will be made in later pages. In the case of Lake Inawasiro-ko, an acid-water lake in Hukusima Prefecture, it was observed that the strong acid-water of River Nagase-gawa (pH: 2.8) flows into the lake and into a somewhat deeper layer than the surface layer and thus lake-water near the river mouth shows the lowest pH-value at the depth of 12.5 meters (YOSHIMURA, 1938 b).

With regard to the inversion of the pH-value in the bottom layer, or the dichotomous stratification, YOSHIMURA and MIYADI published their opinions. According YOSHIMURA (1936 b) it may be due to the increase of the buffer-action in anaerobic bottom water, especially in the lakes whose water shows a very weak buffer-action as is the case with most Japanese lakes.

d) Dissolved Oxygen

With regard to dissolved oxygen detailed data were obtained and are shown in Table II and III. The saturation percentage of oxygen given in the tables was calculated according to FOX (Whipple and Whipple), and the correction of altitude was made according to LAPLACE.

In the present observation the oxygen content increases in the metalimnion and diminishes from this downwards, becoming free at the depth of 9 or 10 meters; particularly so on Aug. 13, 1938, when there occurred a marked supersaturation at the depth of 5 meters (Fig. 5). In the summer of 1934, a similar stratification had been observed by YOSHIMURA, but on the other hand, in the summers of both of 1931 and 1932 the stratification of oxygen was scarcely recognized. It may be possible that this change was caused by the change of the thermal condition of the lake-water mentioned in the proceeding page.

It has been already reported that the maximum amount of oxygen content is found in the metalimnion during the summer stagnation period (JUDAY and BIRGE, 1932; YOSHIMURA, 1938 a). Concerning this fact YOSHIMURA (1935) has given an explanation as follows: the amount of oxygen increases towards the surface by the assimilation of phytoplankton, owing to the better light conditions at the epilimnion in the period of the spring circulation of the lake-water, but the rise in water temperature at the surface in the summer reduces the solubility of O_2 , resulting in supersaturation of oxygen at the surface, and the excess of it evaporates by diffusion. We also discussed in detail the relationship between the transparency of the water and the layer of maximum amount of oxygen content. According to him such a type of stratification of dissolved oxygen as seen in Lake Osore-san-ko is usually observed in the lakes belonging to the oligo- and mesotrophic types of the second and third orders (1938 a). JUDAY and BIRGE (1932) gave some computations with reference to the saturation values of many lake-waters in Wisconsin following MAUCHA's formulae. As a result of the computation, it is also shown that the excess of dissolved oxygen in the thermocline may be due to the assimilation executed by the phytoplankton. In the present case of Osore-san-ko, however, there are two kinds of inflows feeding the lake; namely, the cold water saturated with oxygen which comes from the surrounding mountains, invading into the metalimnion, and the warm water which is derived from the hot-springs spreading over the surface. Thus we must consider the influence of these two kinds of inflow in addition to the assimilation executed by the phytoplankton.

e) Chemical Constituents

The analysis of chemical constituents of this lake water was made by YOSHIMURA using the samples obtained in 1932 and 1934 (TAMURA, 1935:

YOSHIMURA, 1934 a). The results shown in Table IV are those estimated by MATUYA and by the writer in 1936.

TABLE IV. Aug. 4, 1936

Station	Depth m	Cl mg/L.	SiO ₂ mg/L.	P-P ₂ O ₅ mg/L.	N-N ₂ O ₅ mg/L.
St. I.	0	18.1	11.1	0.005	0.000
	4	17.0	9.4	0.022	0.011
	8	16.7	8.2	0.010	0.004
	10	20.6	8.5	0.021	0.008
	14	28.0	15.2	0.125?	0.010
St. II a.	0		13.9	0.007	0.003
	b.		16.3	0.010	0.005
St. III.	0		11.4	0.009	0.000
	5.5		11.5	0.011	0.003
St. IV.	0.5		12.9?	0.008	0.003

St. I: Lake centre

St. II a: Mouth of Tamogizawa (an inlet on the south coast

b: 10 meter upperstream

St. III: off the hotel.

St. IV: Syôzu-gawa

The analysis were made by Z. MATUYA except Cl

Cl: From Table IV and Fig. 6, it can be seen that a slight diminution is observed in the middle layer and a marked increase in the bottom layer. A similar increase of Cl in the bottom water was observed by

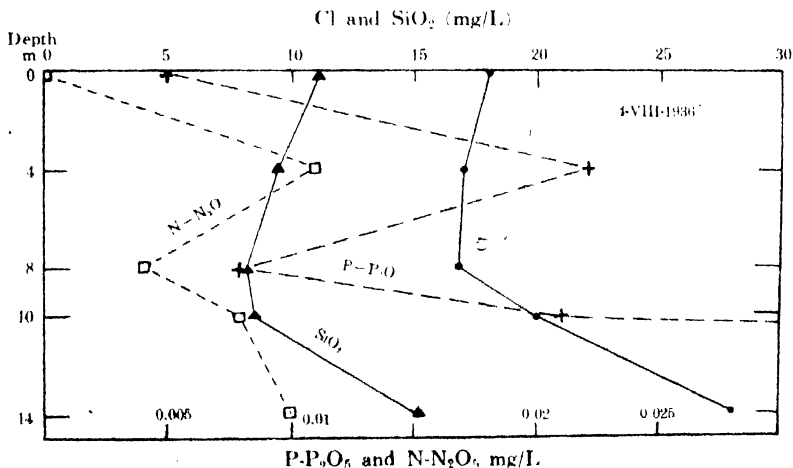


Fig. 6. Chemical constituents of the water of Lake Osoresan-ko.

Ordinate Depth in meter.

Abcissa (upper).... mg/l of Cl and SiO₂.

Abcissa (below).... mg/l of P-P₂O₅ and N-N₂O₅.

YOSHIMURA in the summer of 1934, though the decrease of the same in the middle layer was very slight. According to the studies made by YOSHIMURA (1933) and OHLE (1934) the apparent stratification of Cl indicates the presence of underground water having some different physico-chemical properties.

SiO_2 , $\text{P-P}_2\text{O}_5$, $\text{N-N}_2\text{O}_5$: The data shown in Table IV and Fig. 6 are those estimated by MATUYA by means of colorimetry.

SiO_2 -content is highest in the bottom water and is next in the surface water, and is at the minimum in the water 8 meters deep. $\text{P-P}_2\text{O}_5$ and $\text{N-N}_2\text{O}_5$ are at the minimum in the surface water and increase towards the depth of 4 meters, and again decrease at 8 meters and then become maximum in the bottom water.

It was reported by YOSHIMURA (1934a) that the stratification of Fe showed a similar feature in 1934 as follows:

Depth (m)	0	5	10	12	15
Fe (mg/L)		0.4	0.3	0.2	4.5

According to his investigation, the presence of any marked stratification of iron and of manganese are not to be found in any other acidotrophic lakes of Japan, Lake Osoresan-ko being the only exception (YOSHIMURA, 1936a).

YOSHIMURA found in his investigation of 1934 that the electrical conductivity becomes strikingly reduced from the surface to the bottom. He considered at first that this reduction was caused by the reduction of salinity. But he revised his opinion in his later paper (1936a), adopting OHLE's opinion (1934), that the change of conductivity is mainly due to the decrease of H^+ , a large quantity of sulphuric acid flowing into the surface water of the lake from the hot-springs.

It has been already mentioned that there are two kinds of inflow, namely, a number of small streams of high temperature coming from the hot-springs, and the streams of low temperature coming from the surrounding mountains. Besides these, the marked increase of chloride in the bottom water may indicate the presence of an inflow of underground water of different properties. Thus we are able to consider inflows of at least three kinds in the period of the summer stagnation. Namely, first, warm water flowing into the surface layer, secondly the cold water coming from the mountain and submerging into the metalimnion, and thirdly the underground water remaining in the bottom layer. The anomalous stratification of pH found in the early summers of 1936 and 1938 may possibly be due to this fact.

It is not yet clear by what cause the summer stagnation did not occur in 1931 and 1932. It is, however, reasonable to suggest that the water level might have been higher in those two years than in the later years. If it was so, the water of the mountain streams might have been enabled to mix with the surface water of the lake before submerging into the deeper layer, because the shallow area occupying the margin of the lake was enlarged by the elevation of water level. Hence, it might result in the losing of at least one factor in accelerating the stagnation of the lake water. In fact, according to TAMURA's report the water temperature of the River Ôtukusi-gawa, the largest inflow into the lake, was as high as that of the surface water of the lake, even at a point situated 100 meters distant above the river mouth, and beyond that point it became colder; viz., the water temperature measured at a point 100 meters distant from the river mouth was 23.8° and that of 200 meters was 14.8° on Aug. 13, 1931 (TAMURA, 1933).

III BIOLOGICAL OBSERVATIONS

a) Outline of Ecology

In spite of the strong acidity of the lake-water the littoral region of the lake excepting the north coast is luxuriant in vegetation. As the main littoral plants, we may enumerate *Menyanthes trifoliata* L., *Sparganium ramosum* HUDS. var. *stirabufrym* GRAEBN., and *Scirpus lacustris* L. var. *Tabernaemontani* TRANTS. In the shallower bottom the following subwater forms are found: *Potamogeton nipponicus* MAKINO, *Carex scabrifolia* STEND, and *Leptodictium* sp. (KOKUBO et al, 1931). The last species is found all over most of the area of the lake bottom.

Of animal life *Leuciscus hakonensis* is only the fish inhabiting this lake, its catch amounting to a big quantity. It is said that besides the above species attempts have been made to rear several other fishes such as carp, eel, trout, and crucian in this lake, but all the efforts were in vain. KOKUBO and others (1931) observed that the carp and the crucian did not survive longer than 24 hours in this lake water. According to them, the food contained in the stomach of *Leuciscus* was found to be constituted mostly of benthic Crustacea, *Asellus* sp. which may be rather rarely found in the net collection.

In the small streams coming from the hot-springs as well as in beach water of the lake are found a large number of *Chironomus* larva and the tubes they make. They are most dense in the lake in water of from 30°C

to 34°C. Two species of *Chironomus* are found in this lake. Viz: *Ch. connectens*, and *Ch. plumosus*. In the streams running near the hot-spring, Sukayu, Mt. Hakkôda, Aomori Prefecture, the larva of *Chironomus acerbiophilus* TOKUNAGA is found in a condition closely allied. They live in water of 18°C to 37°C in temperature and is from 3.0 to 3.5 of pH.

b) Plankton

The collection of plankton were made by means of a wing-pump (KOKUBO, 1933), filtering the water with nets made of MÜLLER's gauze No. 25 or of Japanese bolting silk.

The following forms have been found in this lake by TAMURA and the writer. Some forms which are not really planktonic, were also met with.

As zooplankton we may mention the following forms: *Eucyclops serrulatus* (S. FISCHER)? *Macrocyclus fuscus* (JURINE), Copepodid, Nauplius: *Alona* sp., *Chydorus sphaericus* O. F. MÜLLER, *Simocephalus vetulus* (O. F. MÜLLER); *Brachionus urceus* (L.), *Cathypna* sp., *Diaschiza* sp., *Pompholynx* sp., *Tricocerca* sp.; *Chironomus* larva, *Ascellus*, sp., *Hydrocarina*, Nematoda.

Among phytoplankton were noticed, *Eunotia* sp., *Cosinodiscus* sp., *Fragilaria* sp., *Melosira italica*, *Navicula* spp., *Ulothrix* sp., *Anabaena* sp.

Brachionus was the leading form in 1931, and *Simocephalus vetulus* during the period extending 1931-1933, while *Macrocyclus fuscus* has been the leading form since 1934 up to the present time. The phytoplankton seems to have been rather poor in general throughout all these periods. Therefore, in the writer's investigation, it was not dealt with quantitatively. A kind of diatom, *Eunotia* is also found in other acid-water lakes, such as Lake Itibisinai-ko, Gosikinuma group of lakes of Bandai. TAMURA reported a copepod, *Eucyclops serrulatus* in his first paper (1933) but he corrected the name to *Macrocyclus fuscus* in his later paper (1936). It may be, however, possible that *Eucyclops serrulatus* appears occasionally in certain seasons.

A Rotifer, *Brachionus urceus* (L.) (= *B. urceolaris* O. F. MÜLLER) is found sometimes in abundance in this lake. It is known that this species is able to accommodate the pH in a wide range. According to SKADOWSKI (1936) (after RYLOV, 1935, p. 62) the pH-limit ranges from 4.5 to 11, the optimal being 7.6-10. And it was found by the survey made by Viscount TANAKA and others that this species is present in abundance in Lake Itibitinai-ko, Kunasiri Island, the water of which showing such a

pH-value as 2.8. It is of interest to note that the species *B. urceus* is found dominantly in the markedly acid-water in Japan.

It should also be noticed that the season of the maximum abundance of this species in Lake Osoresan-ko seems to vary with the years. For instance, in 1932 it was abundant in July and was very poor in August and September, while in 1934 it was abundant from the end of August to September. In August of 1935 it was somewhat poor in quantity; and in 1936, it was very rare at the end of June, but showed a remarkable dominancy in August. In 1938 it was comparatively poor in quantity in August.

Chydorus sphaericus seems to be a rather common plankton appearing in every collection. This species is typically a littoral form, and a large quantity of it is found in the collection made in the River Syôzu-gawa (Fig. 10 and Table VIII). It was found by UÉNO in Lake Onuma-ike, Nagano Prefecture, the lake water being 3.7 of pH. A Cladoceran, *Simocephalus vetulus*, was the most dominant species in Lake Osoresan-ko in 1931 and 1932, and in 1933, this species appeared still more abundantly. In 1934, however, a striking decrease in quantity was noticed of this species, and the same condition continued until the summer of 1938. The species which has now taken the position of *Simocephalus vetulus* is *Macrocylops fuscus*, a large copepod coloured a beautiful green. This species which had been seldom met with in the years before 1933, has shown a remarkable increase sincethen.

As regards the cause this apparent phenomenon it needs some special comments. With regard to the pH-value and the surface temperature of the lake-water, no noticeable change was recognized, at least not in such a degree as to cause changes in the plankton fauna. It is, however, worthy of attention that the change of plankton above mentioned was accompanied by a marked change of the stratification of the lake-water in summer. In other word, it seems that the stagnation of the lake-water exercised in some way an influence upon the breeding of *Simocephalus vetulus*.

The course of the change in main plankton in the year above mentioned is shown in Fig. 7. As the collections were not always made quantitatively, only the percentage of plankton is given, the phytoplankton being excluded.

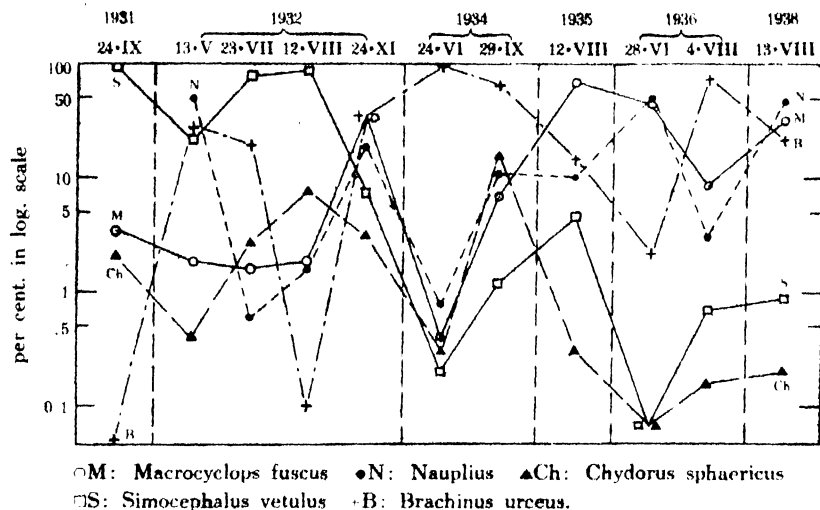


Fig. 7. Feature of changes of the zooplankton from 1931 to 1938, showing the percentage of each plankton. (1931: KOKUBO et al.; '32, '34: TAMURA; '35, '36, '38: MASIKO.)

c) Vertical Distribution and Migration of Plankton

The observation were carried out on June 28-29, on Aug. 4-5, 1936 and on Aug. 13, 1938, making the collection by means of a pump. In the two collections made in 1936, twenty litres of water were obtained from every layer of three meters from the bottom to the surface, and in the collection tried in 1938 ten litres were secured from each of six layers. In 1936 the collections were made four times in the twenty four hours, viz., in the daytime, just after sunset, at night, and at dawn.

The results obtained on June 28-29, 1936, are shown in Table VI and Fig. 8. The main plankton obtained on these days were copepodid and nauplius of *Macrocyclus fuscus*. It was found that the majority of both forms inhabit the thermocline throughout the daytime, but that migration in a slight degree may occur in the twilight and in the night (Fig. 8).

A kind of Rotifer, *Brachionus urceus* was generally found to be distributed in the upper layer, showing its maximum abundance in a layer three meters deep, except in the daytime of the 28th when it was seen on the actual surface. The other Rotifers, such as *Cathypna* sp., *Diaschiza* sp., and *Tricocerca* sp., were generally found in the deeper layer; especially, *Diaschiza* sp. which was densely abundant in a layer 9 meters below the surface or in the part lower than the thermocline.

TABLE V. Showing the vertical distribution of plankton (represented by number of individuals per 10 litres), June 28-29, 1936

Date		14 h. 28/VI						19 h. 28/VI					
Depth (m)	0	3	6	9	12	Aver.	0	3	6	9	12	Aver.	
<i>Macrocylops fuscus</i>	0	0	2	9	0	2.2	4.5	5	5	0.5	0	3	
Copepodid (<i>M. fuscus</i>)	3	4	24	643	4	135.6	7	8.5	8	1000	0.5	204.8	
Nauplius (<i>M. fuscus</i>)	4	30	76	785	0	179.0	58	30	18	1232	0	267.6	
<i>Chydorus sphaericus</i>	0	3	0	0	0	0.6	0	0	0	3	0	0.6	
<i>Simoccephalus retulus</i>	0	0	0	0	0	0.0	2	0	0	0	0	0.4	
<i>Brachionus ureus</i>	54	6	1	0	0	12.2	14	20	2	7	0	8.6	
<i>Cathypna</i> sp.	2	3	2	4	0	2.2	0	1	1	2	0	0.8	
<i>Diasthiza</i> sp.	0	0	0	12	0	2.4	0	0	2	10	0	2.4	
<i>Pompholyx</i> sp.	0	1	1	3	0	1.0	1	0	0	0	0	0.2	
<i>Tricocerca</i> sp.	3	3	1	8	0	3.0	5	1	5	5	0	3.2	
Eggs	58	20	3	3	0	16.8	7	9	2	3	0	4.5	
Nematoda	0	0	0	0	0	0.0	0	0	0.5	0	0	0.1	
Total	124	70	110	1457	4	355.0	98.5	74.5	43.5	2262.5	0.5	496.0	

Date		21 h. 28/VI						5 h. 29/VI					
Depth (m)	0	3	6	9	12	Aver.	0	3	6	9	12	Aver.	
<i>Macrocylops fuscus</i>	0.5	5.5	4.5	4	0	2.9	3	6	10.5	0.5	0	4.6	
Copepodid (<i>M. fuscus</i>)	13	21	86	2201	0	464.2	31	40	39	660	0.5	154.2	
Nauplius (<i>M. fuscus</i>)	15	12	44	1910	0.5	396.7	31	50	58	1172	0	262.2	
<i>Chydorus sphaericus</i>	0	0.5	0	1	0	0.3	0	0	0	0.5	0	0.1	
<i>Simoccephalus retulus</i>	0	0	1	0	0	0.2	0	0.5	1.5	1.5	0	0.7	
<i>Brachionus ureus</i>	11	32	0	1	1	9.0	9	42	27	0	0	13.0	
<i>Cathypna</i> sp.	1	2	1	9	0	2.6	4	2	0	2	0	1.6	
<i>Diasthiza</i> sp.	0	0	3	2	2	1.6	0	0	2	4	0	1.1	
<i>Pompholyx</i> sp.	0	0	0	0	0	0.0	1	0	0	0	0	0.2	
<i>Tricocerca</i> sp.	0	1	4	10	2	3.4	1	1	4	2	0	1.6	
Eggs	18	18	1	1	1	7.8	2	5	17	0	0	4.8	
Nematoda	0	0	0.5	0	0	0.1	0	0	0	0	0	0.0	
Total	59.5	92	145	4139	8.5	888.8	82.5	145.5	159	1845	0.5	446.7	

TABLE VI. Showing the vertical distribution of plankton (represented by number of individuals per 10 litres), Aug. 4-5, 1936

Date	15:30' 4/VIII (clear)							18:30' 4/VIII (dull)						
	0	3	6	9	12	14	12	0	3	6	9	12		
<i>Macrocylops fuscus</i>	0	2.5	8.5	18	0	0	0	5	10	15.5	11	0		
Copepodid (<i>M. fuscus</i>)	4	4.5	10	16.5	0.5	0	0	5.5	8	28.5	19.5	0.5		
Nauplius (<i>M. fuscus</i>)	28	14	7	1.5	0	0	0	18.5	2	6	1.5	0.5		
<i>Cydorus sphaericus</i>	1	1	0.5	0.5	0	0	0	0.5	0	0	2	0		
<i>Simocephalus vetulus</i>	6	2.5	4	1	0	0	0	3.5	2	2	1	0		
<i>Brachionus urceus</i>	1247	321	25.5	3	2	2.5	2	399	6	7.5	10	9.5		
<i>Cathypna</i> sp.	0	0.5	0	0.5	0.5	0	0	0	0	1	0.5	0		
Eggs	84	44	1	2	0	0.5	0	151.5	2	3.5	7	3		
Total	1370	390	56.5	43	3	5.5	3	586.5	30	64	52.5	13.5		

Date	21:31' 4/VIII (foggy)							6 h 5/VIII (rain)						
	0	3	6	9	12	14	14	0	3	6	9	12		
<i>Macrocylops fuscus</i>	18.5	16	7.5	12.5	0	0	0	5.5	3.5	3	6	0		
Copepodid (<i>M. fuscus</i>)	16	14	27	42	0	1	5	5	7	13	12	0		
Nauplius (<i>M. fuscus</i>)	10.5	8.5	4	1	0	1.5	8	8	8	10	1.5	0.5		
<i>Cydorus sphaericus</i>	0	0	0.5	0	0	0	0	0.5	0	0	1	0		
<i>Simocephalus vetulus</i>	0.5	1	2	0.5	0	0	0	0.5	3	3	0.5	0?		
<i>Brachionus urceus</i>	498	330	22	7	6	32?	122	314	73.5	0.5	10.5			
<i>Cathypna</i> sp.	1	0	0.5	0.5	0	0	0	0	0	0	0.5	0		
Eggs	54.5	67.5	3.5	1.5		10.5	36	18.5	7	0.5	0.5	0.5		
Total	599	437	67	64	6	45	177.5	354	109.5	14.5	11.5			

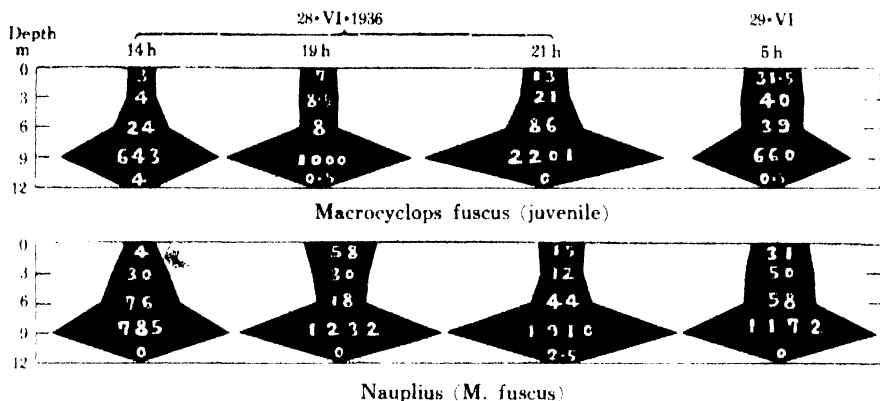


Fig. 8. Vertical distribution of juvenile and nauplius of *Macrocyclus fuscus* on June 28-29, 1936 (Numbers of individuals per 10 litres of water are given).

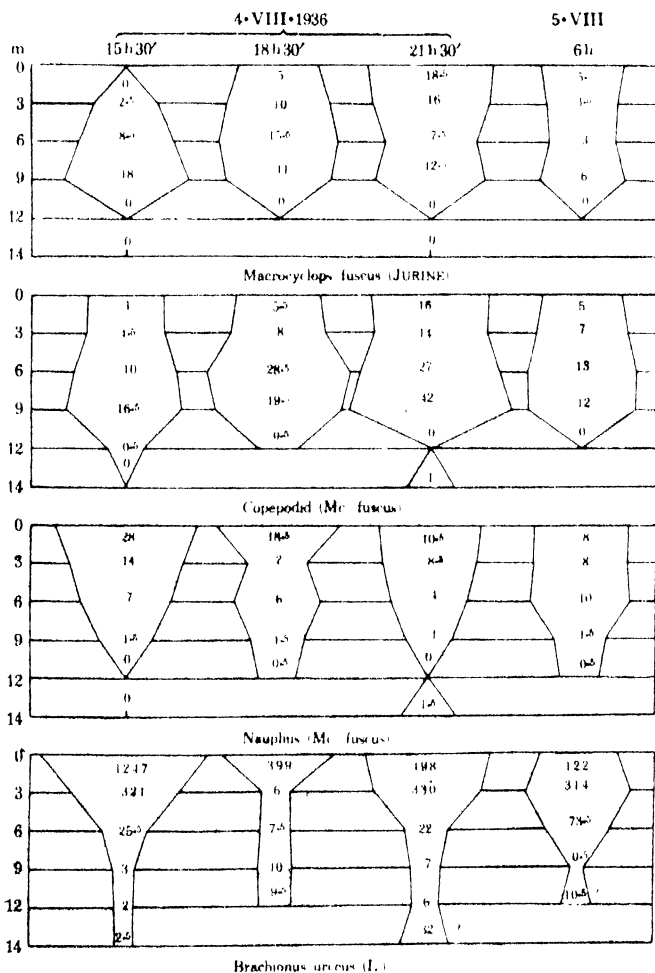


Fig. 9. Vertical distribution of main plankters on Aug. 4-5, 1936. (Numbers of individuals per 10 litres of water are given).

The results obtained on Aug. 4-5, 1936, are shown in Table VI. The features of the diurnal migration of the main plankton, namely those of the adult, juvenile, and nauplius of *M. fuscus* and of *B. urceus* are shown in Fig. 9. As is seen from the table, the adult form of the former became somewhat dominant, but the total numbers of the adult and of the juvenile were much less when compared with those seen in the former observation. The adult was observed to increase in number during the night in the surface water, from which it was completely absent in the daytime, during which time it appeared most abundantly in the thermocline. The juvenile also showed a similar upward movement in the night. Its upward movement was in slighter degree than in the case of the adult, but it was far more noticeable when compared with the migration of the juvenile in former times. The nauplius appeared most abundantly in the surface layer in the daytime, showing a sharp contrast to the former, except in the early morning, when the distribution was nearly uniform from the surface down to a depth of 6 meters. The species *Brachionus urceus* mostly inhabited the very surface. A slight decrease in the maximum number seen in the early morning may possibly have been due to the heavy rain which fell that morning. A similar tendency was recognized also in the observation made in 1936. It is, however, not definitely certain if the vertical distribution of this kind of Rotifer was influenced by the rain-fall or by the wind as in the cases of some phytoplankton, indicated by STADTMANN (RUTTNER, 1914).

TABLE VII. Vertical distribution of the plankton (represented by the number of individuals per 10 litres). 15 h., Aug. 13, 1938

Depth (m)	0	2	5	8	10	13	Aver. (%)
<i>Macrocyclops fuscus</i>	0	3	7	2	0	1	2.2 (2.9)
Copepodid (<i>M. fuscus</i>)	31	45	19	23	2	6	21.0 (28.2)
Nauplius (<i>M. fuscus</i>)	103	60	22	9	1	7	33.7 (45.2)
<i>Chydorus sphaericus</i>	0	1	0	0	0	0	0.2 (0.2)
<i>Simoecephalus vetulus</i>	3	0	0	1	0	0	0.7 (0.9)
<i>Brachionus urceus</i>	39	21	19	4	3	10	16.0 (21.5)
Eggs	2	1	0	2	0	0	0.8 (1.1)
Total	178	131	67	41	6	24	74.5 (100.0)

The results obtained on Aug. 13, 1938, are shown in Table VII and Fig. 10. The adult form of *Macrocyclops fuscus* showed its maximum

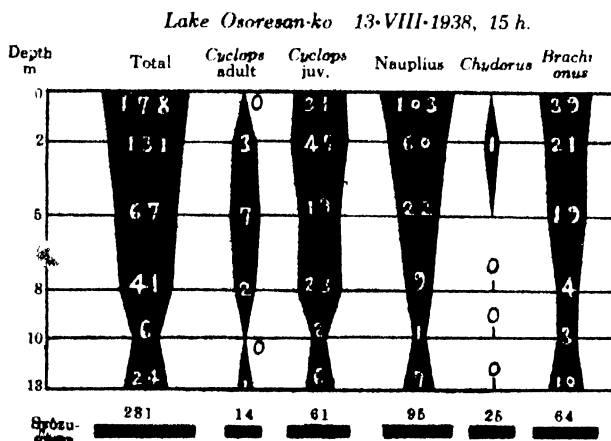


Fig. 10. Vertical distribution of plankton on Aug. 13, 1938 (Numbers of individuals per 10 litres are given).

number in the layer 5 meters deep, and was nearly completely absent in the surface layer, and only one specimen was found in the water 13 meters deep where no oxygen was detected. The juvenile of this species was found most abundantly in the layer 2 meters deep, and next most abundantly in the surface layer. The nauplius of the same species crowded mostly in the surface layer, especially in the uppermost part. *Brachionus urceus* was found to be distributed also in the deeper water, though it also may densely inhabit the very surface.

In general consideration of the vertical distribution of the plankton some interesting facts were noticed. The adult of *Macrocyclus fuscus* always inhabits the thermocline during the daytime, and migrates in the night. The juvenile of the same species inhabits the metalimnion, showing more or less a tendency to migrate upwards in the night or in the twilight. The nauplius of the same species shows a much different distribution according to the season. On June 28-29, 1936, most of it was inhabiting the thermocline, in August of 1936 and 1938 it inhabited mainly the surface layers. Thus the vertical distribution of *Brachionus urceus* seems to vary according to the year or to the season, the diurnal migration being scarcely recognized. According to TAMURA, this species inhabited a comparatively deep layer in the summer of 1932, while in the autumn of 1934 it inhabited rather the upper layer. In the summer of 1936, the writer found this species mostly inhabiting the surface water during the daytime, and the same condition was also obtained in 1938, but it was not so distinct as in 1936.

The fact that the whole mass of plankton was found somewhat in the deeper layer on Aug. 13, 1938, may be regarded as having been caused by the heavy fog on that day. This fact was also observed by RUSSEL (1926) and KIKUCHI (1930).

In 1936, *Macrocyclus fuscus*, both the adult and juvenile forms, crowded just above the anaerobic layer where the proportion of dissolved oxygen was only 53.5 and 45.5 percent, on June 28 and on Aug. 4 respectively. On the other hand, in 1938 they were found in the uppermost layer of the thermocline where the oxygen was in a state of oversaturation.

According to KIKUCHI's observation made in 1931 and in Lake Hange-tu-ko, some plankton, such as *Pseudodiaptomus japonicus*, *Limnocalanus sinensis*, *Leptodera kindtii*, *Keratella cochlearis* inhabited abundantly the layer of scanty oxygen. The vertical distribution of *Polyarthra platyptera* observed by RUTTNER in Lunzer Obersee is also one of the remarkable examples. In this lake almost all the individuals of that Rotifer were found to be crowded in the layer where the O_2 -content was only 0.1 mg per litre (RUTTNER 1914). The vertical distribution of nauplii in Lake Mendota shows features similar to those observed by the writer in Lake Osore-san-ko on June 28, 1836 (JUDAY 1903, quoted from RUTTNER 1914).

Thus we meet with many examples which serve to show the fact that some plankton often prefer to live in the water-layer of poor oxygen content.

IV. PLANKTON IN THE WATER FLOWING FROM THE LAKE THROUGH RIVER SYÔZU-GAWA

Through the only outlet, Syôzu-gawa, a large quantity of plankton is found in the water flowing from the lake. The writer has tried two collections in the outlet above mentioned, i) obtaining the plankton from 10 litres of water taken by means of a pump on Aug. 13, 1938; ii) obtaining the same by filtering the water for 30 seconds by means of a net of 20 cm diameter on Aug. 14. The results thus far secured are as shown in Table VIII.

When comparing the above table with Tables V-VII, one will be aware that of every form of plankton the number of individuals contained in an equal volume of water is much greater in the water which came from the outlet than in the water taken from lake centre. If we assume that the section area of the river is roughly thirty times as large as the mouth area of the net, the number of individuals which passes into the river

TABLE VIII. Showing the plankton obtained by the collection tried in the River Syôzu-gawa

plankton	No. of individuals per 10 L. (%)		No. of ind in 30 sec. collec. (%)	
<i>Macrocyclops fuscus</i>	14	5.2	1066	14.0
Copepodid	61	22.5	1312	17.2
Nauplius	95	35.1	869	11.1
<i>Chydorus sphaericus</i>	25	9.2	1831	24.1
<i>Simocephalus vetulus</i>	2	0.8	10	0.13
<i>Brachionus urceus</i>	64	24.0	2233	29.3
<i>Cathypna</i> sp.	0	0.0	53	0.7
<i>Duschnia</i> sp.	0	0.0	2	0.03
Eggs	10	3.7	206	2.7
<i>Asellus</i> sp.	0	0.0	1	0.01
<i>Chironomus</i> larva	0	0.0	5	0.07
Nematoda	0	0.0	18	0.24
Total	281	100.5	7609	99.88

from the lake may be estimated at as much as 2,191,392 a day.

In the summer of 1932 MORI (1933) made some quantitative observation on the plankton of Syôzu-gawa. In his case the species predominating in the lake was *Simocephalus vetulus* and consequently the river plankton also consisted of the same species. His results showed that the number of plankton decreases with the course of the river, thus, after running a course of 11 km the number of plankton became 16% of the initial quantity, being measured at the Pacific estuary. Of all the species the decrease of *S. vetulus* was the most remarkable, probably owing to the delicate structure of its body.

V. ANORGANIC ACIDOTROPHIC LAKES IN JAPAN

The acidotrophic lake-type is divided by YOSHIMURA into the following two subtypes:

- i) Organogene or organic acidotrophic type: the acidity of the lake water originates from the humic substance.
- ii) Mineralogene or anorganic acidotrophic type: the acidity of the lake water is caused by inorganic acid, chiefly by sulphuric or hydrochloric acid, having its origin in active volcanoes or hot-springs.

In our country where the volcanoes and the hot-springs are very common it is quite natural that many lakes are of the latter type. The strongest

acidotrophic lake hitherto known in the world is Lake Kata-numa in Miyagi Prefecture (YOSHIMURA, 1934 b). No plankton can be found in this lake, but some benthic forms were reported recently by HUZIMATU (1938) and by NEGORO (1938 a). They are *Pinnularia Braunii* (GRUN.) var. *amphi-cephala* (A. MAYER); *Stylonichia* sp., *Callidina bidens* GOSS, as species of Notommatidae, *Chironomus aceriphilus* TOKUNAGA, Ch. sp. (Salinarius-group), *Forcipomya* sp., a species of belonging to Ephyridae. Besides these there were two adult forms of insect, *Enochrus umbratus* SHARP and *Aquarius palludum* FABRICIUS collected by HUZIMATU, but he considered them as temporary migrates from elsewhere. Next to Lake Kata-numa comes Lake Okama of Zaô which is one of the representative crater lakes

TABLE IX. Showing the general features of the anorganic acidotrophic lakes found in Japan

Lake	Locality (Pref.)	Dep. m	pH	Litt. Plant	Benthos			Plankton	Fish	Investigator
					Moss	Diatom	Animal			
Kata-numa	Miyagi	22	1.4-1.5	—	—	+	+	—	—	1), 2)
Okama (Zaô)	Miyagi	43	1.9-3.4	—	—	—	—	—	—	3)
Onuma-ike	Nagano	26	2.8-3.7	—	—	—	—	+	—	4)
Itibisinai-ko	Kurile	62	2.8	+	—	—	—	+	—	5)
Hudô-ike	Miyazaki	9	2.9	+	+	+	—	—	—	6)
Osorezan-ko	Aomori	16	3.0-4.0	+	+	+	+	+	+	
Aka-numa (Bandai)	Hukusima	4	3.3-3.6	+	+	—	—	—	—	7), 8)
Ruri-numa	Hukusima	9	4.3	+	+	+	•	+	+	8)
Ao-numa	Hukusima	6	4.4	+	+	+	•	+	+	8)
Bisayamon-numa	Hukusima	13	4.5	+	+	+	+	+	+	8)
Benten-numa	Hukusima	6	4.5-4.7	+	+	+	•	+	+	8)
Inawasiro-ko	Hukusima	93	4.4-5.4	+	+	+	+	+	+	9), 10)
Aka-numa (Hokkôda)	Aomori	18	4.7	+	+	+	+	+	+	6)
Okotanbe-ko	Hokkaidô	21	4.9	+	•	+	+	+	+	11)
Kuttyaro-ko	Hokkaidô	125	5.1-6.0	+	+	+	+	+	+	12), 13)

1) YOSHIMURA (1934 b)

2) HUZIMATU (1938)

3) ANZAI (1934)

4) UENO (1934 b)

5) TANAKA & HOSHINO (1934)

6) YOSHIMURA (1937 a, 1937 b)

7) YOSHIMURA (1935 b)

8) YOSHIMURA, NEGORO & YAMAMOTO (1936)

9) YAMAMURA (1934)

10) YOSHIMURA (1938 b)

11) TAKAYASU & IGARASHI (1935)

12) TAKAYASU, IGARASHI & KONDÔ (1933)

13) MASIKO (1936)

laying near the summit of Volcano Zaô, showing as high pH as from 1.9 (1931) to 3.4 (1933) (ANZAI, 1934). No organisms were found in this lake. The lake habitable for plankton is Lake Itibisinai-ko of Kunasiri Island, and its pH is uniformly 2.8 from surface to bottom (TANAKA and HOSHINO, 1934). In this lake *Brachionus urceus* was found abundantly (i.e. 99% all of the plankton in the lake) and *Keratella cochlearis* (GOSSE) and *Chydorus gibbus* LILLJEBORG were also found but only meagrely, no phytoplankton being found (UENO, 1934 a). Of a pond, called Itibisi-syôko which lies near L. Itibisinai-ko, it is reported that its water has a pH as high as 2.3 and a temperature higher than 7°C. It contains no oxygen at all even in the surface layer and thus is, lacking of organisms (TANAKA and HOSHINO, 1934). Lake Ônuma-ike is also known as one of the marked acidotrophic lakes (pH; 2.8-3.4). This lake is inhabited but scanty by such creatures as *Bosmina longispina*, *Chydorus sphaericus*, and *Cyclops strenus* (UENO, 1934 b). Taking the above lakes into consideration, Lake Osoresan-ko may be said to be the most acid lake in which the fish and phytoplankton are found.

The general feature of anorganic acidotrophic lakes in Japan is tabulated in Table IX.

VI. ACKNOWLEDGEMENT

In carrying out the present work the writer has obtained guidance, first from Prof. Dr. S. HATAI and afterwards from Prof. Dr. S. HÔZAWA of the Tôhoku Imperial University. It is a great pleasure to express here the writer's sincere gratitude to them for their kindness. The writer wishes to express his hearty thanks to Dr. S. KOKUBO for his kind guidance and advice given in the course of preparing the paper. To Mr. T. TAMURA and to Dr. S. YOSHIMURA, the writer is indebted for valuable advice given during the work. Further the writer must express his gratitude to Mr. Z. MATUYA and Mr. T. TEZUKA who have kindly put their unpublished data at his disposal,

VII. SUMMARY

1) Lake Osoresan-ko is an extreme example of an acidotrophic lake. If more particular mention is required it may be said to belong to the class of lakes of mineralogene or anorganic acidotrophic subtype. The pH-value of the lake water ranges around 3.5.

2) The productivity of this lake seems to be fairly great in spite of

its high acidity. The lake may be the most acidotrophic lake in the world inhabited by fish and by phytoplankton.

3) Since 1924 the lake water has shown some distinct stratification in summer, being observed by both physical and chemical means, while before that in the summer of 1931 and 1932 the stratification was scarcely recognized. A similar change was also observed in the case of the transparency of the water.

4) We may consider the inflow of water into the lake to be of at least three kinds of categories, viz., i) the water derived from the hot-springs, ii) the river water coming from the surrounding mountains, and iii) the underground water indicated by the Cl-content.

5) The species of plankton from Lake Ososesan-ko hitherto recorded are about twenty five in number. During the period covering from 1931 to 1933, the lake was predominated by a cladoceran, *Simocephalus vetulus*, which species has shown a marked decrease in number from 1934 to 1938. The form which increased thereafter was a copepod, *Macrocyclus fuscus*, replacing the above species. A Rotifer, *Brachionus urceus* also appeared abundantly in the lake. The latter species seems to have a special adaptability to water of high acidity.

6) The change of plankton above mentioned is simultaneous with and probably caused by change in the stratification of the lake water during the summer stagnation period.

7) The vertical distribution of the main plankton has been observed. *Macrocyclus fuscus* inhabits the thermocline during the daytime, and shows an obvious nocturnal migration. Copepodid showed a similar behaviour though it is less distinct in degree. Nauplius of the same species was mostly collected in the lower part of thermocline during daytime in the early summer of 1936, while it was found largely in the surface layer in the midsummer of that year. *Brachionus urceus* always inhabited the surface layer, while some other rotifers, especially *Diaschiza* sp., collected at the thermocline.

8) Concerning the outflow of plankton from the lake through the river, an observation was made quantitatively of the River Syôzu-gawa, the only outlet of Ososesan-ko.

REFERENCES

- ANZAI, T., 1934: Limnological Study on the Crater Lake of Volcano Zaô, after Ceasing of its Volcanic Action (in Japanese). Proc. Saitô Hô-on Kai, Nos. 85, 86.
HUZIMATU, K., 1938: On the Aquatic Organisms found in Lake Kata-numa, a Very Strong

- Acid-water Lake in Japan (in Japanese with Eng. résumé). *Ecol. Rev.*, Vol. 4, p. 131.
- JUDAY, C. and E. A. BIRGE, 1932. Dissolved Oxygen and Oxygen Consumed in the Lake Waters of Northeastern Wisconsin. *Trans. Wisconsin Acad. Sci. Arts Lettr.*, Vol. 27, p. 415.
- KIKUCHI, K., 1930: Diurnal Migration of Plankton Crustacea. *Quart. Rev. Biol.*, Vol. 5, p. 189.
- , 1932: Influence of Dissolved Oxygen upon the Vertical Distribution of Zooplankton in Lake Suigetsu (in Japanese). *Japanese Jour. Limnol.*, Vol. 1, p. 30.
- KOKUBO, S., 1933: A Plankton Pump in which the Winding of the Tow Line of Section Tubing is made Automatically (in Japanese with Eng. résumé). *Bull. Japanese Soc. Sci. Fish.*, Vol. 1, p. 277.
- , T. TAMURA and N. ABE, 1931: Biological Study on Lake Osorezan (in Japanese). *Proc. Saitō Hō-on Kai*, No. 59.
- MASIKO, K., 1934: Observations on Lake Akan-ko and Lake Kuttyaro-ko in Summer Time (in Japanese). *Japanese Jour. Limnol.*, Vol. 4, p. 136.
- , 1936: Measurement of the Light Intensity penetrated into Waters off Asamusi and at Lake Osorezan-ko by means of Hydriodic Acid (in Japanese with Eng. résumé). *Bull. Japanese Soc. Sci. Fish.*, Vol. 5, p. 189.
- , 1938: Plankton Studies on Lake Osorezan-ko (in Japanese with Eng. résumé). *Japanese Jour. Limnol.*, Vol. 8, p. 45.
- MCCREA, R. H., 1923: Light Intensity Measurement by means of Hydriodic Acid. *Jour. Ecol.*, Vol. 11, p. 103.
- MORI, K., 1933: On the Plankton of River Syōzu-gawa (in Japanese). *Proc. Saitō Hō-on Kai*, No. 74.
- NEGORO, K., 1938: A Species of Diatom living in the Bottom-mud of Lake Kata-numa, a very Strong Acid-Water Lake in Japan (in Japanese). *Botany and Zoology*, Vol. 6, p. 1731.
- OHLE, W. 1934: Chemische und physikalische Untersuchungen nord-deutscher Seen. *Arch. f. Hydrob.*, Vol. 26, pp. 386, 584.
- ŌISHI, M., 1935: Biology of Chironomus larva (in Japanese). *Ecol. Rev.*, Vol. 1, p. 45.
- PEARSALL, W. H., 1921: The Aquatic Vegetation of the England Water. *Jour. Ecol.*, Vol. 8, p. 163.
- RUSSEL, F. S., 1926: The Vertical Distribution of Marine Plankton. IV. *Jour. Mar. Biol. Assoc., N. S.*, Vol. 14, p. 415.
- RUTTNER, F., 1914: Die Verteilung des Plankton in Süßwasserseen. *Forts. Naturwiss. Forsch.*, Vol. 10, p. 273.
- RYLOV, W. M., 1935: Das Zooplankton der Binnengewässer. *Die Binnengewässer*, Stuttgart.
- TAKAYASU, S. and H. IGARASHI, 1935: Preliminary Report of the Investigation of Lake Okotanbe (in Japanese). *Japanese Jour. Limnol.*, Vol. 5, p. 117.
- TAKAYASU, S., H. IGARASHI and K. KONDO, 1933: Investigation of Lake Kuttyaro-ko (in Japanese). *Rep. Fish. Research (Hokkaidō)*, Vol. 28.
- TAMURA, T. 1933: Preliminary Report of Research of Lake Osoreyama (in Japanese). *Japanese Jour. Limnol.*, Vol. 2, p. 76.
- , 1936: Plankton of Strongly Acid Water Lake Osorezan-ko (in Japanese). *Japanese Jour. Limnol.*, Vol. 6, p. 63.
- TANAKA, A. and R. HOSHINO, 1934: Limnological Survey of the Lakes in the South-western Part (South of 44° n. lat.) of Kunasiri-sima (in Japanese). *Japanese Jour.*

- Limnol. Vol. 3, p. 95.
- UÉNO, M., 1934 a: Plankton of the Lake in the Island of Kunasiri (in Japanese). Japanese Jour. Limnol., Vol. 3, p. 129.
- , 1934 b: Acid Water Lakes in North Sinano. Arch. f. Hydrob., Vol. 27, p. 571.
- YAMAMURA, Y., 1934: Distribution of the Hydrogen Ion Concentration in the Water of Lake Inawasiro (in Japanese). Japanese Jour. Limnol., Vol. 3, p. 50.
- YOSHIMURA, S., 1932: Annual Change of the Transparency of Lake Tazawa-ko (in Japanese). Japanese Jour. Limnol., Vol. 2, p. 15.
- , 1933: Chloride as Indicator in Detecting the Inflowing into an Inland-water Lake of Under-ground Water, Possessing Special Physico-chemical Properties. Proc. Imp. Acad., Vol. 9, p. 156.
- , 1934 a: Anohaline Stratification of the chemical Constituents of Lake Osorezan-ko, Aomori Prefecture, Japan. Proc. Imp. Acad., Vol. 10, p. 475.
- , 1934 b: Kata-numa, a Very Strong Acid-Water Lake on Volcano Katanuma, Miyagi Prefecture, Japan. Arch. f. Hydrob., Vol. 26, p. 197.
- , 1935: Relation between Depth for Maximum Amount of Excess Oxygen during the Summer Stagnation Period and the Transparency of Freshwater Lakes of Japan. Proc. Imp. Acad., Vol. 11, p. 356.
- , 1936 a: Contributions to the Knowledge of Iron dissolved in the Lake Waters of Japan. 2nd Rep. Japanese Jour. Geol. Geogr., Vol. 13, p. 39.
- , 1936 b: Dichotomous Stratification of pH in Lake Waters (in Japanese). Kagaku, Vol. 6, p. 508.
- , 1937: Characters of Anorganic Acidotrophic Lakes in Japan (in Japanese). Nippon Gakujutukyôkai Hôkoku, Vol. 12, p. 161.
- , 1938 a: Dissolved Oxygen of the Lake Waters of Japan. Sci. Rep. Tôkyô Bunrika Daigaku, Ser. C, No. 8, p. 64.
- , 1938 b: Sounding and Hydrological Condition of Lake Inawasiro-ko. Geogr. Rev. Japan, Vol. 14, p. 532.
- , K. NEGORO and S. YAMAMOTO, 1936: Limnological Reconnaissance of the Gosikinuma Lake Group of Volcano Bandai, Hukusima Prefecture (in Japanese with Eng. résumé). Geogr. Rev. Japan, Vol. 12, p. 1.

ON THE OCCURRENCE OF THE ACID REACTION IN THE BODY TISSUES OF AN ASCIDIAN, *ASCIDIA SAMEA* OKA¹⁾

By

SATARÔ KOBAYASHI

Zoological laboratory, Hakodate College of Fisheries, Hakodate, Hokkaido, Japan

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INTRODUCTION

Since HENZE's work (1911, 1912, 1913 a and 1913 b) on the presence of free sulphuric acid in the blood-cells of *Phallusia mammillata* and in the fluid of the mantle-tissue of *Ascidia mentula*, a number of work have appeared by several other investigators.

HECHT (1917) demonstrated an acid reaction in the blood-cells, but not in the plasma, of the Atlantic ascidian, *Ascidia atra* by testing it with litmus paper.

Histochemically RAPKIN and DAMBOVICEANU (1925), when observing the blood of *Ascidia mentula* using the staining reaction with pH indicators and by the microinjection technique, determined the pH value of the plasma-fluid and the intracellular pH of the blood-cells and also of the vesicular cells in the test.

PRENANT (1925) in his studies of the acid or alkaline reaction in the test in a certain number of the European ascidians by means of CLARK and LUBB's indicator series, detected the acid reaction in the test of several species, viz.: *Perophora listeri*, **Diprosoma listrianum*, *Ascidia fumigata*, and also in the blood of **Leptoclinum gelatinosum*.

WEBB (1939) made observations of the blood, using a number of the European ascidians, with special reference to the biochemistry of the vanadium chromogen. From the results of his observation and reexamination of the earlier works, he states that in *Ascidiiidae* and *Perophoridae*, sulphuric acid is presented in the special type of the blood-cell, the

¹⁾ Contribution from the Marine Biological Station, Asamushi, Aomori-Ken, No. 169.

* According to WEBB's paper (1939), the species given by PRENANT (1925) as *Diplosoma listerianum* correspond to *Leptoclinum gelatinosum*, and *Leptoclinum gelatinosum*, to *Didemnum lahiliei* HARTM.

vanadocyte, yet in some form within Ascidiidae, such as *Ascidia aspersa*, and *Ascidia scabra*, acid was found in the vesicular cell of the blood as well as in the test. In addition to the above species so far studied, he ascertained the presence of sulphuric acid in both the test and also the blood of *Ascidia conchilega* var. *depressa*, *Ascidia aspersa* and *Ascidia scabra*.

A condition similar to that of the above ascidian species was reported by the present writer as occurring in the blood-fluid of the Pacific ascidian, *Chelyosoma siboja* OKA (KOBAYASHI 1933, 1935 and 1938); free sulphuric acid was also demonstrated in the blood-fluid, a considerable amount of acid was found to have accumulated in the special corpuscle cell existing among the various different corpuscle cells, i.e. in the cell sap of the vesicular type cell. Chemical analysis of the blood-fluid showed that the amount of sulphuric acid (ca. 4%) in the corpuscle-fluid is nearly identical with the isotonic saline concentration of the surrounding sea-water.

In 1939, the present writer had an opportunity to study the acid reaction in the body-tissues of *Ascidia samea* OKA, by the courtesy of Mr. MASAHICO TAKAMATSU, of the Saito Ho-on Kai Museum in Sendai.

The species under consideration was recently described as *Ascidia samea* by Dr. ASAJIRÔ OKA in his report on the simple ascidians of Mutsu Bay (OKA 1935). He reported that this species is distributed along the Pacific coast of north-eastern Honshu Japan. During the writer's stay at Ishijima village on Katsura-jima, an islet in Matsushima Bay near Sendai, Miyagi prefecture, it was found that *Ascidia samea* OKA was in great abundance on the rocky shore of that islet. These ascidians were attached to the rock surface by the entire left side of the body, together with other sessile animals and sea weeds.

The medium sized specimens measured 3.5–5.0 cm. in length, 2.0–2.5 cm. in breadth and 1.0–2.0 in thickness.

A thin cartilaginous test 0.5–2.0 mm. in thickness covers the outer layer of the entire soft body.

In this species, the acid reaction was also demonstrated in the living tissues when litmus paper was applied on a cut surface of the ascidian body, as in the case of *Chelyosoma siboja* OKA.

This work was undertaken in the summer 1939 to determine the nature, the amount and the anatomical localization of acid present in the ascidian body in *Ascidia samea* OKA in comparison with that in *Chelyosoma siboja* OKA.

The preliminary treatment for *Ascidia samea* was carried out in the

laboratory of the Biological Institute, Tōhoku Imperial University in Sendai, and that for *Chelyosoma siboja* OKA at the laboratory of the Asamushi Marine Biological Station.

After this preliminary treatment, the materials of both samples were taken to the zoological laboratory of this college where this work was continued. The results so far obtained are presented in this paper.

MATERIAL AND METHOD

The materials employed in this experiment, were collected at the low tide in the summer season, being carefully removed from the rock surface.

The materials thus collected were cleansed of their adhering mud and other sessile organisms. Of these sample materials, a number were dissected, in order to obtain the body-fluid and tissue-fluid for chemical analysis of the acid in them.

Others of the specimens were used for the detection of acid reaction in the living tissue by means of vital and other dyes.

For the chemical analysis of the body-fluid and of the tissue-fluid from the soft body wall, two groups were selected from the samples. Those of first group were respectively dissected dorsoventrally, so that the outer cartilaginous test could easily be separated from the ascidian soft body. In this treatment it was found that the body-fluid (presumably the blood-fluid) oozed out like a pool in the region between the test and the soft body. From the soft ascidian body thus treated, the tissue of the actual soft body-wall was obtained, after the removal of the internal organs. It was minced, collected in a small flask and then heated on a boiling water bath for 10 min. By this treatment the tissue-fluid of the soft body wall was obtained. It was filtered and used for chemical analysis. On the other hand, from a large number of the second group, about 100 cc. averaging 1.5–2.0 cc. for each individual was collected by a pipet. It was immediately centrifugalized and separated into the plasma-fluid and the corpuscle was sedimented at 3000 r.p.m. for 20 minutes. The corpuscle-fluid was obtained by freezing from the sedimented corpuscle, and this was further centrifugalized and filtered as in the case of *Chelyosoma siboja* OKA. (KOBAYASHI 1933, 1935 and 1938).

For the chemical analysis of the ascidian tissue-fluid, the acidity of the plasma-fluid and of the frozen corpuscle-fluid, was determined with 0.1 N. sodium hydroxide solution by phenolphthalein and pH was measured by the hydrogen ion meter by means of the hydrogen electrode.

Protein was removed by precipitating with the method of FOLIN and WU (1919) or by 20% solution of trichloroacetic acid. Chlorine was estimated by the method of WHITEHORN (1921) or VORHARD, and inorganic sulphate by the gravimetric method of barium chloride. The treatment and method of the chemical analysis of these ascidian-fluids were almost the same as in the blood-fluid of *Chelyosoma siboja* OKA. (KOBAYASHI 1933, 1935 and 1938).

The body-fluid of *Chelyosoma siboja* OKA, and the specimens of the sea-water from the habitat of both the ascidians, (*Ascidia samea* OKA and *Chelyosoma siboja* OKA) were analysed for comparison with those in the case of *Ascidia samea* OKA.

For the staining reaction of acid, neutral red, methyl red and aethyl-violet were employed. Neutral red was dissolved in the filtered sea-water, in which a number of the living ascidians were then immersed for two days. Methyl red and aethylviolet were employed under the microscopic examination for the detection and the determination of the cellular pH in the acid cells for which the latter was specially chosen for the low pH indicator in the range of pH 1.00-0.00, instead of methyl-violet in the case of *Chelyosoma siboja* OKA (KOBAYASHI 1938).

The violet-coloured testing-paper prepared by aethylviolet dye, was used for the detection of acid reaction after the removal of the outer cartilaginous test, it was applied on the surface of the soft ascidian body and also in the body-fluid.

EXPERIMENTAL RESULTS

Chemical analysis of the tissue-fluid of the soft body wall and the body-fluid.

The analytical results of the tissue fluid of the soft body wall and of the body-fluids (plasma-fluid and corpuscle-fluid) are shown in Table 1, together with that of the Matsushima sea-water.

TABLE 1.

		Acidity		Gm. in 1000 cc.		SO ₄ /Cl
		pH	Normality	Cl	SO ₄	
Tissue fluid of soft body wall		1.27	0.248	7.24	21.44	2.4680
Body-fluid (Blood-fluid)	Plasma-fluid	2.28	0.015	15.20	4.18	0.2292
	Corpuscle-fluid	0.58	0.581	2.10	37.44	14.7629
Matsushima sea-water		8.0	—	16.44	2.31	0.1166

As shown in the above table, the pH of the ascidian fluids were respectively determined to be 1.27 for the tissue-fluid, 2.28 for the plasma-fluid and 0.58 for the frozen corpuscle-fluid. The total acidity or normality was estimated to be 0.248 for the tissue-fluid, 0.015 for the plasma-fluid and 0.581 for the corpuscle-fluid. The amount of chlorine (gm. in 1000 cc.) was determined to be 7.24 for the tissue-fluid, 15.20 for the plasma-fluid, 2.10 for the corpuscle-fluid, while the amount of inorganic sulphate was found to be 21.44 for the tissue-fluid, 4.18 for the plasma fluid and 37.44 for the corpuscle-fluid. The results of the Matsushima sea water analysis were found to be 8.0 for pH, 16.44 for the chlorine content and 2.31 for sulphate.

The ratios of SO_4/Cl were respectively found by calculation to be 2.4680 in the tissue-fluid of the soft body wall, 0.2292 in the plasma-fluid, 14.7629 in the corpuscle-fluid and 0.1166 for the Matsushima sea-water.

From the results of the above chemical analysis of the ascidian fluid, the values of the acidity (normality and pH) and the amount of inorganic sulphate were found to be the greatest in the corpuscle-fluid, lowest in the plasma-fluid, and of medium value in the tissue-fluid, while chlorine content was found to be greatest in the plasma-fluid, lowest in the corpuscle-fluid and of medium value in the tissue-fluid.

However, the sulphate content and the ratios of SO_4/Cl in the Matsushima sea-water show the lowest value, while pH and Cl content show the highest values as compared with those in the ascidian fluids.

From the above results, it is evident that the acid reaction of the ascidian tissue-fluid and body-fluid in *Ascidia samea* OKA is due to the presence of free sulphuric acid and that the acid concentration is highest in the corpuscle-fluid.

Staining reaction by the vital and other dyes.

After keeping the ascidians for 24-48 hours in a glass bowl containing the filtered sea-water in which neutral red had been dissolved, 5-10 cc. of the body-fluid could be collected. By allowing this fluid to stand for several hours in a test tube, it was observed that it separated into two portions, i. e. the supermanent plasma-fluid and the sedimented corpuscle mass. The upper layer of the plasma-fluid exhibited a weak acid reaction, in marked contrast to the heavy deep wine-red colour of the strong acid reaction in the sedimented corpuscle mass.

On examining this separated corpuscle mass under the microscope, the enormous volume and multitudinous number of the vesicular cells and

a certain number of the vesicular amoeboid cells were readily visible, being stained by the pink colour of the acid reaction at the vacuolated cell sap. Besides these two types of acid cells, other types of non-acid cells which lack the acid reaction are also present in this fluid. The diameter of these cells were measured and found to be 40-60 μ for the vesicular cell, 8-30 μ for the vesicular amoeboid cell, most of which were 8-13 μ , and 5-7 μ for the other remaining non-acid cells.

The general morphological characteristics of the vesicular type cells and other non-acid cells in the body-fluid of *Ascidia samea* OKA show much similarity to those in the blood of *Chelyosoma siboja* OKA, as was observed by OHUYE (1935) and by the present writer (1933 and 1938).

By the staining reaction by methyl-red, it was observed under the microscope, that the cell sap of the vesicular cell was stained a deep wine-red colour, indicating a clear evidence of acid, a pH below 4.4 as was employed by HENZE (1912). It was further observed by the staining reaction of aethylviolet under the same conditions that the cell-sap of the vesicular cell was stained a yellowish colour of the high acid concentration of pH 1.00-0.00, and yet the external plasma-fluid surrounding the vesicular cell gradually stained a greenish clour, indicating a lower acidity than the cell-sap of the vesicular cell.

Similar acid reaction produced by aethylviolet in the cell-sap of the vesicular cell does not only show *in vivo* under the microscope but also in the frozen corpuscle-fluid *in vitro*, which closely agrees with the results obtained from the frozen corpuscle-fluid (pH 0.582) when employing the electrometric method.

It was further noted that no cellular elements such as the "Balasenzellen" could be detected in the cartilaginous test of this species.

A further experiment on the acid reaction was performed on the soft body surface immediately after the removal of the cartilaginous test.

On applying the aethylviolet tesing paper on the surface of the soft body-wall, it was found that the proper deep violet-colour of the testing paper was suddenly changed into the yellowish-green colour of the acid reaction, showing slightly less acidity than the frozen corpuscle-fluid. This colour-change of acid reaction appeared as many scattered spots of the coloured reaction over the whole surface of the ascidian soft body, due to the locarization of the presence of acid.

In *Chelyosoma siboja* OKA, a similar experiment with relation to the coloured reaction by aethylviolet paper was also carried out on the soft body-surface after the removal of the cartilaginous test, and it was found

that there is no evidence of colour-change or acid-reaction in any part of the soft body except the clear acid colour-reaction at the blood sinus, located at the ventral side of the ascidian body, as was described in the previous paper (KOBAYASHI 1938). The experiment on *Ascidia samea* OKA suggest that a considerable number of the acid-containing vessels are distributed on the surface of the soft body-tissue, showing a close anatomical connection between the test and the soft body. In addition to this fact, in the fixed specimen preserved in the BOUIN's picro-formal solution or formaline solution, the distribution of fine branching blood vessels was observed all over the surface of the soft body, and on the other hand, in the cartilaginous test, the blood vessels are considered to be anastomosingly distributed through those on the soft body surface.

GENERAL REMARKS

From the results obtained by this experiment, the ascidian, *Ascidia samea* OKA, shows the acid-reaction in the soft body-tissue and in the body-fluid which oozed out through the soft body tissue and the cartilaginous test. No acid reaction of the cellular element however could be detected in the cartilaginous test, as in the case of *Chelyosoma siboja* OKA.

The acidity concentration of sulphuric acid is found to be high (3%) in the corpuscle-fluid, low (0.7%) in the plasma fluid and of medium value (1.2%) in the tissue-fluid of the soft body-wall.

As to the occurrence of acid in the plasma-fluid, RAPKIN and DAMBOVICEANU (1925), after the separation of the blood-cells by centrifugalization gave a pH of 7.0-7.4 in the European species, *Ascidia mentula*.

WEBB (1939) claims in his experiment with *Ascidia aspersa* and other ascidians that the plasma of these ascidians is usually found to be close to pH 7.0 by careful centrifugalization. In connection with the above works, the present writer has also observed that the vesicular cells in both *Ascidia samea* OKA and *Chelyosoma siboja* OKA, are easily cytolysed either by mechanical or other shocks, such as centrifugalization, owing to the very delicate thin cell-membrane of the vesicular cell.

It is very difficult to obtain the non-acid containing plasma-fluid or to avoid the rupture of the vesicular cells in the plasma-fluid without any artificial mechanical disturbance, at least, at the presence of such special type of the cells in the ascidian body.

Comparing these results obtained from the body-fluid in *Ascidia samea* OKA with those in *Chelyosoma siboja* OKA, as will be shown in Table 2,

the acidity amounting to SO_4 and Cl , and also the ratios of SO_4/Cl in *Ascidia samea* respectively show a lower value than those in *Chelyosoma siboja*. And the difference in these two species are found to be much greater in the corpuscle-fluid introduced from the vesicular acid cells, than in the plasma-fluid.

In connection with above differences in the analytical value between these two ascidian species, *Ascidia samea* and *Chelyosoma siboja*, the Cl and SO_4 content in the Matsushima Bay sea-water show considerably less value than in the Mutsu Bay sea-water, although both show almost similar values in the ratio of SO_4/Cl .

Previous to this experiment, YOSHIMURA and NISIYAMA in their oceanographical study observed in 1937 the low salinity of the water in Matsushima Bay.

TABLE 2.

Species	Acidity		Gm. in 1000 cc.		SO ₃ /Cl
	pH	Normality	Cl	SO ₄	
Plasma-fluid					
<i>Ascidia samea</i> OKA	2.28	0.015	15.20	4.18	0.2292
<i>Chelyosoma siboja</i> OKA	1.80	0.027	17.99	4.96	0.2297
Corpuscle-fluid					
<i>Ascidia samea</i> OKA	0.58	0.581	2.10	37.20	14.7619
<i>Chelyosoma siboja</i> OKA	0.38	0.876	2.14	45.65	17.7787
Sea-water					
Matsushima Bay	8.0	---	16.44	2.31	0.1166
Mutsu Bay	8.2	---	19.14	2.67	0.1161

The above results in the low concentration in acid (sulphuric acid and pH) as well as in the amount of Cl and SO_4 in both the plasma and corpuscle-fluid shown by *Ascidia samea* OKA, are considered to be isotonically affected by the low salinity of the surrounding sea-water in Matsushima Bay in comparison with those shown by *Chelyosoma siboja* OKA.

Microscopically it was found that free sulphuric acid is contained in the cell sap of the vesicular type cell, chiefly in such cells with a diameter

of 40–60 μ . These vesicular type cells morphologically show a striking resemblance to those in the blood of *Chelyosoma siboja* OKA (OHUYE 1935 and KOBAYASHI 1933, 1938).

In the blood of *Ascidia mentula*, RAPKIN and DAMBOVICEANU (1925) determined the cellular pH of the ascidian corpuscle, using bromphenol-blue and thymolblue, and by the microinjection technique. They observed that among certain types of the blood-cells, both the adipophore and the vesicular element are acid containers, showing a pH below 2.8.

Other types of the blood-cells gave the higher pH value alkaline than pH 6.0, against the pH 8.1 of the sea-water, while the large vesicular cell or bladder-cells embedded in the tunic show an acid reaction indicating a pH nearly 2.0.

In the writer's own experiment in *Ascidia samea* OKA by means of aethylviolet, it was found that the cellular pH in the cell-sap of the vesicular cell gave the value between pH 1.00–0.00, agreeing with a pH 0.582 obtained by the electrometric method in the frozen corpuscle-fluid. This figure in pH in the cell-sap of the vesicular cell in *Ascidia samea* OKA shows resemblance to that in *Chelyosoma siboja* OKA (pH 0.38) (KOBAYASHI 1938).

Anatomically the body-fluid presumably originates from a certain number of the blood-vessels distributed on the surface of the soft body-tissue, which is continuous to the anastomosing blood-channels in the cartilaginous test.

Such anatomical connection through a large number of blood-vessels greatly differs from the connection through only one blood-sinus at the ventral side of the soft body in *Chelyosoma siboja* OKA (KOBAYASHI 1938).

It may therefore be concluded that the chief source of the acid reaction in *Ascidia samea* OKA is due to the presence of free sulphuric acid accumulated in the cell sap of the vesicular type cells in the blood-fluid.

Such a type of the occurrence of the vesicular cell was reported by WEBB (1939) in the European species, *Ascidia aspersa* and *Ascidia scabra* and by the present writer in *Chelyosoma siboja* OKA (KOBAYASHI 1933, 1935 and 1938).

But *Ascidia samea* OKA is different from the European species in the fact that there is no acid reaction in the test, and that it rather resembles that in *Chelyosoma siboja* OKA.

SUMMARY

Ascidia samea OKA shows the acid reaction in the body-fluid which oozed out from the cartilaginous test, and the soft body-tissue. No acid-cellular element could be detected in the cartilaginous test.

Vesicular cells of enormous volume and in multitudinous numbers, are found in the body-fluid of *Ascidia samea* OKA, and they closely resemble those in the blood of *Chelyosoma siboja* OKA. Sulphuric acid is accumulated in the cell sap of the vesicular type cell which reaches 3% or pH 0.528.

The acid concentration in the vesicular cell-sap in *Ascidia samea* OKA is affected by the low salinity in the surrounding sea-water of Matsushima Bay.

Anatomically a certain number of the blood-vessels distributed over the whole surface of the soft body, connect both the test and the soft body.

The acid reaction in *Ascidia samea* OKA, seems chiefly due to the presence of sulphuric acid in the vesicular type cell contained in the body-fluid (presumably the blood).

The occurrence of acid reaction of a type similar to that in *Ascidia samea* OKA is found in the European species, *Ascidia aspersa* and *Ascidia scabra* (WEBB 1939). But there is acid reaction in the tests of these two European species, which in this point differ from *Ascidia samea* OKA, the latter more closely resembling *Chelyosoma siboja* OKA.

REFERENCES

1. FOLIN, O. and WU, H. 1911. A Sytem of Blood Apalysis. Journ. Biol. Chem., Vol. 63, pp. 80-110.
2. HECHT, S. 1917. Physiology of *Ascidia atra* LEUSEUR. Amer. Journ. Physiol., Vol. 45, pp. 157-187.
3. HENZE, M. 1911. Untersuchungen über das Blut der Ascidien. I. Zeitsch. F. Physiol. Chem., Vol. 79, pp. 494-501.
4. HENZE, M. 1912. Untersuchungen über das Blut der Ascidien. II. ibid., Vol. 79, pp. 215-228.
5. HENZE, M. 1913. a) Untersuchungen über das Blut der Ascidien. III. ibid., Vol. 86, pp. 340-344.
6. HENZE, M. 1913. b) Über das Vorkommen freier Schwefelsäure im Mantel von *Ascidia mentula*. ibid., Vol. 86, pp. 345-346.
7. KAHN, B. S. and LEIBOFF, S. L. 1928. Colorimetric Determination of Inorganic Sulphate in Small Amount of Urine. Journ. Biol. Chem., Vol. 80, pp. 277-285.
8. KOBAYASHI, S. 1933. Studies on Acid of the Body Fluid from an Ascidian, *Chelyosoma siboja* OKA. Sci. Rep., Tōhoku Imp. Univ., Biol., Vol. 8, pp. 277-285.

9. KOBAYASHI, S. 1935. Chemical Composition of the Body Fluid of an Ascidian, *Chelyosoma siboja* OKA. Sci. Rep. Tôhoku Imp. Univ. Biol., Vol. 9, pp. 407-413.
10. KOBAYASHI, S. 1938. Studies on the Body Fluid of an Ascidian, *Chelyosoma siboja* OKA with Special Reference to its Blood System. Sci. Rep., Tôhoku Imp. Univ., Biol., Vol. 13, pp. 25-35.
11. OHUYE, T. 1935. On the Coelomic Corpuscle in the Blood of some Japanese Ascidian. Sci. Rep., Tôhoku Imp. Univ., Biol., Vol. 11, pp. 191-206.
12. OKA, A. 1935. Report of the Biological Survey of Mutsu Bay 28. Ascidian Simplicies. Sci. Rep., Tôhoku Imp. Univ., Biol., Vol. 10, pp. 427-446.
13. PRENANT, M. 1925. Contribution à l'étude cytologique du calcaire II. sur les condition de formation des spicule chez didemnides. Bull. Biol., Vol. 59, pp. 403-453.
14. RAPKIN, L. et DAMBOVICEANU, A., 1925. Sur le pH intérieur de certains éléments du sang et la tunique chez *Ascidia mentula*. Compt. Rend. Soc. Biol., Vol. 93, pp. 1427-1429.
15. WEBB, D. A. 1939. Observation on the Blood of Certain Ascidians, with Special Reference to the Biochemistry of Vanadium. Journ. Exp. Biol., Vol. 16, pp. 499-523.
16. WHITEHORN, J. C., 1921. A System of Blood Analysis. Journ. Biol. Chem., Vol. 45, pp. 449-560.
17. YOSHIMURA, S. and NISIYAMA, S. 1937. Oceanographical Observation of Matusima Bay, Miyagi Prefecture in the Winter of 1937. Proc. Imp. Acad. Tokyo, Vol. 13, pp. 103-106.

RELATIVE GROWTH OF THE CRAB, *SESARMA* (*HOLOMETOPUS*) *DEHAANI* M. EDWARDS

By

IKUSÔ HAMAI

East Asia Institute, Surugadai, Tôkyô

AND

ETURÔ HIRAI

Biological Institute, Tôhoku Imperial University, Sendai

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Up to the present time, much information has been gathered regarding the relative growth of Crustacea, and some interesting facts have been disclosed with special reference to growth gradient and to sex phenomena. From these data the nature and features of growth have been discussed. Many of these investigations are analysed by the method of the application of the allometric formula. This method is certainly effective in the analysis of relative growth in some cases, but it is difficult to apply in the study of Crustacea, as it is liable to be inaccurate in the observations of moultings and in the determinations of instars of which size-measurements were to be made. This fact has been pointed out by HAMAI (1937) with regard to other animal-groups, *e. g.* Mollusca, in which the age of the animals is an important factor in the analysis of relative growth besides the size of the creatures, as in the case of the moulting in Crustacea.

The present investigation constitutes an addition to our knowledge of the relative growth and sex dimorphism in Crustacea, examining the correlation between the relative growth and the moultings, as a brief note previous to further detailed investigations. Accordingly, the growth of the carapace has been discussed, and the body-growth as well as that of the abdomen and the chela have been observed for the sexual differences.

Before proceeding further, the writers wish to express their sincerest thanks to Prof. E. NOMURA for his cordial guidance in the course of this investigation, to Assistant Prof. I. MOTOMURA for his kindness in reading the manuscript, and to Dr. T. SAKAI who willingly identified the species.

MATERIAL AND METHOD

Sesarma (Holometopus) dehaani is a common crab in shallow fresh water in Japan. The material specimens were collected in July and August during which time they are in the reproductive period, from the paddy fields of Dotahuti and the neighbouring region in Isinomaki.

Measurements were made of specimens preserved in a 75% solution of alcohol. Four linear dimensions were measured: 1) the carapace-width, 2) the carapace-length—the distance from the end of the frontal lobe to the posterior end of the intestinal region, 3) the abdomen-width—the greatest width of the third abdominal segment, which shows most remarkable differences in the two sexes, and 4) the chela-length—the maximum length of the propodite of the right chela. The right and left chela are almost the same size.



Fig. 1. Photograph of mature *Sesarma (Holometopus) dehaani*. Right: male, and left: female. $\times \frac{1}{2}$

The total number of specimens examined were 366. Three sets of measurements, viz. the carapace-width and the carapace-length, the carapace-width and the abdomen-width, and the carapace-width and the chela-length, were respectively made independently in both sexes. Accordingly, 48.75 individuals were used in the calculation of each of six sets. The method of the calculation is that of the least squares in the logarithms of respec-

tive measurements in the equation of the allometry, $\log y = \alpha \log x + \log b$, in which x is the carapace-width as the standard, and y another dimension comparable with x , α and b being the constants representing the equilibrium constant of relative growth or the differential growth ratio (HUXLEY and TEISSIER, 1936) and the local index (HAMAI, 1939) respectively, plotting individually these measurements. The probable errors of α and $\log b$ were also calculated.

RESULTS

In male crabs the log-log plots of the respective set of measurements show breaks and discontinuity of the straight line at about 16 mm. carapace-width. Then, two stages of growth are seemingly distinguishable (Table 1 and Fig. 2).

TABLE 1
Values of α and $\log b$

	Stage	α	No.	α	No.	α	$\log b$	P.E.
Carapace-length	I α	1.027 \pm 0.013	18	1.053 \pm 0.009	53	-0.026 \pm 0.016		1.6
	I $\log b$	1.9003 \pm 0.0128		1.8635 \pm 0.0120		0.0368 \pm 0.0175		2.1
	II α	1.087 \pm 0.015	47	"		0.031 \pm 0.017		2.0
	II $\log b$	1.8192 \pm 0.0201		"		-0.0443 \pm 0.0231		1.9
Abdomen-width	I α	1.035 \pm 0.036	19	1.510 \pm 0.023	25	-0.475 \pm 0.043		11.0
	I $\log b$	1.5690 \pm 0.0359		1.2133 \pm 0.0258		0.3557 \pm 0.0442		8.0
	II α	0.938 \pm 0.017	51	1.020 \pm 0.045	30	-0.082 \pm 0.048		1.7
	II $\log b$	1.6958 \pm 0.0222		1.8630 \pm 0.0615		-0.1672 \pm 0.0654		2.6
Chela-length	I α	1.199 \pm 0.031	19	1.155 \pm 0.013	48	0.044 \pm 0.034		1.3
	I $\log b$	1.5588 \pm 0.0310		1.5728 \pm 0.0169		-0.0140 \pm 0.0353		0.4
	II α	1.449 \pm 0.020	56	"		0.294 \pm 0.024		12.3
	II $\log b$	1.2682 \pm 0.0275		"		-0.3046 \pm 0.0323		9.4

The discontinuity of the allometric equation in the carapace-length/width relation, which is shown between the smaller and larger sized groups of male crabs, is very slight, but it may be recognizable as a slight difference of variation which occurs at moulting. It must, however, be further investigated by more observations.

The abdomen-width appears to increase at a lower rate of growth than the carapace-width above from the size of about 16 mm. in carapace-

width comparing with the specimens below this size. But the difference of the two stages is statistically insignificant, and they are necessarily considered as a simple allometry in combining the two, or rather considered as isometrical.

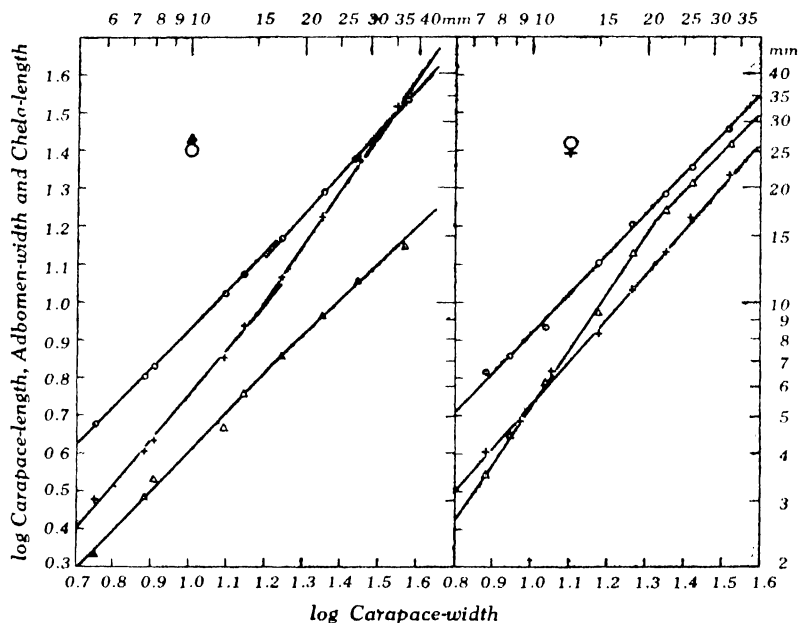


Fig. 2. The log-log plots of the mean of logarithms of various dimensions.
 ○ carapace-length, + chela-length, △ abdomen-width.

The chela-length of the male crabs and the abdomen-width of the females begin to show a different growth-ratio distinctly after a definite size has been attained, viz. in the male chela a higher ratio is attained at the size of about 16 mm. carapace-width, and in the female abdomen the intensive allometry of the smaller sized crabs lowers until an isometric ratio almost parallel with that of the carapace-length after about 20 mm. carapace-width has been reached (Tables 2 and 3).

In the younger stage of the female, the carapace, abdomen, and the chela respectively increase with different growth intensity. The abdomen shows particularly the highest allometry, and accordingly the width of abdomen which was smaller than the chela-length in the crabs below 10 mm. in carapace-width, increases and becomes larger than the chela-length after the attainment of the size of about 10 mm. in carapace-width,

TABLE 2

Differences of α and $\log b$ between the various parts in the males

		Carapace-length	Abdomen-width	Chela-length
Carapace-length	Differences of α	0.060 \pm 0.020	0.008 \pm 0.038	0.172 \pm 0.034
	.. $\log b$	0.0811 \pm 0.0238	0.3313 \pm 0.0381	0.3415 \pm 0.0335
Abdomen-width	Differences of α	0.149 \pm 0.023	0.097 \pm 0.040	0.164 \pm 0.057
	.. $\log b$	0.1234 \pm 0.0299	0.1268 \pm 0.0422	0.0102 \pm 0.0174
Chela-length	Differences of α	0.362 \pm 0.025	0.511 \pm 0.026	0.250 \pm 0.037
	.. $\log b$	0.5510 \pm 0.0341	0.4276 \pm 0.0353	0.2906 \pm 0.0411

The figures in the rectangles enclosed by the thick lines on the diagonal are the differences between the younger and older stages, those above this diagonal show the differences between the various parts in the younger stage, and the figures below this diagonal are of those in the older stage.

TABLE 3

Differences of various α and $\log b$ in the females

		Carapace-length	Abdomen-width	Chela-length
Carapace-length	Differences of α		0.457 \pm 0.025	0.102 \pm 0.016
	.. $\log b$		0.6502 \pm 0.0285	0.2907 \pm 0.0207
Abdomen-width	Differences of α	0.033 \pm 0.046	0.490 \pm 0.051	0.355 \pm 0.026
	.. $\log b$	0.0005 \pm 0.0627	0.6497 \pm 0.0667	0.3595 \pm 0.0308
Chela-length	Differences of α		0.135 \pm 0.047	
	.. $\log b$		0.2902 \pm 0.0638	

This table is also constructed in the same manner as Table 2

viz. during the changes of the chela-length from 51% to 60% of the carapace-width, the abdomen-width increases from 46% to 77%. The male abdomen is isometric and the difference between the two sexes is very remarkable in the younger stage. The relative width of the female abdomen is larger than that of the males also in smaller crabs.

The chela-length is, in both sexes, almost similar in size relative to the carapace-length during the younger stage, and shows, then, an almost similar relation in relative growth, but in the older stage the chela of the male obtains an intensive allometry showing a secondary sexual character, and finally becomes greater than that of the female. As the result of

this intensive allometry, the male chela develops finally to the same size as the carapace-length or more, viz. the male chela-length is 64% of the carapace-width at the size of 16 mm. in carapace-width, while the carapace-length is 81% of the carapace-width at that size, and after the growth, the proportion of the chela at the size of 38.5 mm. carapace-width increases up to 96% of the width of carapace over the carapace-length, which is, almost equal to the former proportion, 86% at that size.

The carapace does not show any significant differences in the sexes throughout either the younger or older stages, and the relative sizes are also almost similar in both sexes. The changes of the form of the carapace are, then, very simple both in the males and the females, except for the slight discontinuity of the male crabs in the length-width relation of the carapace. The growth of the carapace is isometric or slightly positively allometric in both sexes.

DISCUSSIONS

In the investigations of the growth of Crustacea, the size at moulting and the number of moults are necessarily important. But for these observations, confusion would occur in the determination of allometry and direction of size-variation, as pointed out by HAMAI (1937, '38) with regard to the Molluscan shells in which the age and the variation of size have been considered. TEISSIER (1935) has also distinguished the relative growth and the trend of variability in size as "dysharmonie de croissance" and "dysharmonie de taille" respectively. He has considered that by the break and discontinuity of the relative growth curve the existence of the growth stage can be detected, and in each growth stage, the growth is continuous. The critical point dividing two growth stages is supposed to occur with the changes of equilibria in internal secretion or humoral correlation. HUXLEY (1932) has also discussed interesting problems on the allometry and moulting.

NEEDHAM (1937) has discussed the accuracy of the rhythmic change of α stating as follows: "It is not a question of a mere moulting effect in the case of *Asellus*. In the first place, moulting occurs more frequently than the number of waves in the graph of α ; secondly, individual differences in the time of moulting would obscure any such effects on a graph of grouped data, or even on a graph of individuals". In such a case, confusion between the relative growth and the trend of variability may not occur. But in the cases when the number of moultings is small

and the variation in size is large, it is quite possible that confusion may occur.

In the present case, moultings have not been observed, but, because the frequency polygon of the carapace-width shows multi-modality, and is bimodal in the neighbouring region of the breaking point which divides the relative growth curve into two stages, viz. the younger and older stages, each stage accordingly extends over two or more instars accurately. Any allometric equations in *Sesarma* show, then, the relative growth but not the trend of size-variability or "dysharmonie de taille". PAULIAN (1936) has proved in *Eupagurus prideauxii* the discontinuity in the right claw of the males by the fact that in the neighbouring region of this discontinuity the frequency polygons of the claw are clearly bimodal. In this case the relative growth may be distinguished from the trend of variability.

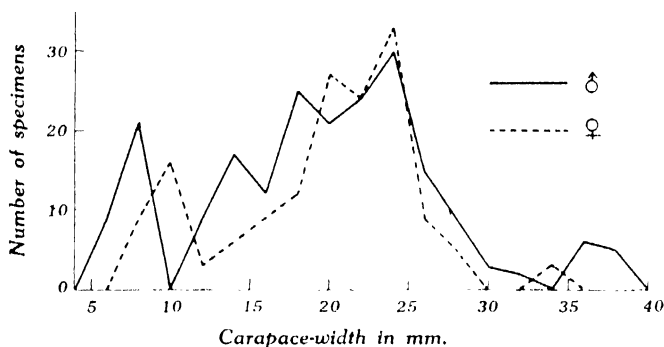


Fig. 3. Frequency polygons of the carapace-width.

GRAY and NEWCOMBE (1938 a) have studied the relative growth of *Callinectes sapidus* expressing the dimensional relations by the equation, $y=mx+b$, where x and y are the comparable dimensions and m and b are the constants. In their case, as the constant b in this equation is not zero, the relative growth is allometric. The data given by them can be accurately enough described by the allometric equation. The consideration by the latter equation might be rather more effective than the former in comparing the relative growth ratio. Here, what is arising in the question, is not dy/dx , but, indeed, $\frac{dy}{y} / \frac{dx}{x}$. Referring to their observations on moultings (1938 b), we have analysed their data as follows (Tables 4 and 5).

TABLE 4
Relative Growth of Callinectes sapidus (Male)

Growth stage and the constants of allometry	Mean carapace-width (mm.)	Mean carapace-length (mm.)	Calculated carapace-length (mm.)	Difference	Remarks
I $\alpha=0.93078$ $\log b=1.80759$	35.55	17.68	17.83	0.15	Including 4 instars at least.
	45.26	22.40	22.32	-0.08	
	54.49	26.50	26.53	0.03	
	65.39	31.72	31.44	-0.28	
	74.28	35.29	35.40	0.11	
	85.35	40.12	40.28	0.16	
II $\alpha=0.92551$ $\log b=1.81711$	93.71	44.21	43.86	-0.35	Including 2 instars at least. Sexual maturity occurs at the beginning of this stage.
	104.61	48.25	48.56	0.31	
	115.31	52.71	53.14	0.43	
	125.29	57.91	57.38	-0.53	
	134.37	60.22	61.22	1.00	
	145.72	65.35	64.60	-0.75	
III $\alpha=0.74825$ $\log b=0.19507$	134.37	60.22	61.32	1.10	Including 2 instars at least. GRAY and NEWCOMBE (1938b) have estimated that as the result of moulting, the carapace-width increases from 126.5 to 155 mm. and from 155 to 184 mm. in the specimens with the initial width 20 mm., and from 135 to 164 mm. and 164 to 195 mm. in the specimens with the initial width 80 mm.
	145.72	65.35	64.04	-1.31	
	154.49	67.77	68.06	0.29	
	163.71	71.42	71.08	-0.34	
	174.68	73.97	74.62	0.65	

As shown in Tables 4 and 5, during the first and second stages the values of α do not indicate any very significant differences, but during the second stage the sizes of the carapace-length in the females are slightly smaller than in the males. In the third stage too, the values of α are almost similar in both sexes, but the percentages of the carapace-length compared with the carapace-width are lower in the females than in the males. In both sexes, the significant drop occurs in the ratio of relative growth rates from 0.93 to 0.75 in male crabs and from 0.90-0.94 to 0.73 in the females. GRAY and NEWCOMBE (1938 a) have stated that in the width range below 140 mm. a close parallelism exists between the males and the females with respect to the length-width and the eye-to-spine-width relations. This conclusion generally coincides with that of our analysis. They have further concluded that above this point, marked variation occurs. In the case of the females, two distinct relations obtain when the length is compared with the width and the eye-to-spine variables. The break occurs at about the 140 mm. point, the average width at which they

TABLE 5
Relative Growth of Callinectes sapidus (Female)

Growth stage and the constants of allometry	Mean carapace-width (mm.)	Mean carapace-length (mm.)	Calculated carapace-length (mm.)	Difference	Remarks
I $\alpha = 0.89882$ $\log b = 1.86343$	36.60	18.37	18.57	0.20	Including 4 instars at least.
	45.34	22.48	22.51	0.03	
	55.29	27.05	26.90	-0.15	
	64.95	31.13	31.09	-0.04	
	74.48	35.10	35.16	0.06	
	84.30	39.19	39.30	0.11	
II $\alpha = 0.94002$ $\log b = 1.77305$	94.34	42.60	42.59	-0.01	Including 2 instars at least.
	104.66	47.25	46.95	0.30	
	113.76	50.13	50.78	0.65	
	124.72	55.44	55.37	0.07	
	132.71	58.98	58.70	-0.28	
III $\alpha = 0.73197$ $\log b = 0.20361$	124.72	55.44	55.47	0.03	GRAY and NEWCOMBE (1938b) have estimated that as the result of moulting, the carapace-width increases from 114 to 157 mm. in the specimen with the initial width 20 mm. and from 108 to 149 mm. in the specimens with the initial width 80 mm. Including 2 instars or 2 at most. # Mature stage with only one instar.
	132.71	58.98	58.06	-0.92	
	145.34	60.59	62.07	1.48	
	155.10	61.82	65.11	0.29	
	165.12	68.51	68.18	-0.33	
	172.71	70.74	70.17	-0.57	
	#				

become mature. In male specimens, increase in width is accompanied by a proportionate increase in the other dimensions. However, in our opinion, the distinct drop in the relative growth ratio occurs in "both" sexes, and the drop in the females occurs at the moulting of maturation. The formula, $L = 7.111 + 0.370 W$, which has been given by GRAY and NEWCOMBE to the specimens above the 140 mm. carapace-width, shows only the trend of variability or "dysharmonie de taille". On the other hand, $\alpha = 0.73$ here calculated indicates the relative growth ratio from premature to mature, approximately. GRAY and NEWCOMBE (1938 b) have considered that at the initial width of about 89 mm., sexual maturity occurs in the males. This occurrence is recognizable as a slight decrease of α from the first (premature) stage to the second (mature) stage (Fig. 4). Thus, although the allometric equation is merely an approximation (HAMAI, 1937), it is clearly effective in cases where the other facts are evident. Further evidences have been given by ANDERSON, LUMER and ZUPANCIC (1937) and WEYMOUTH and MACKAY (1936).

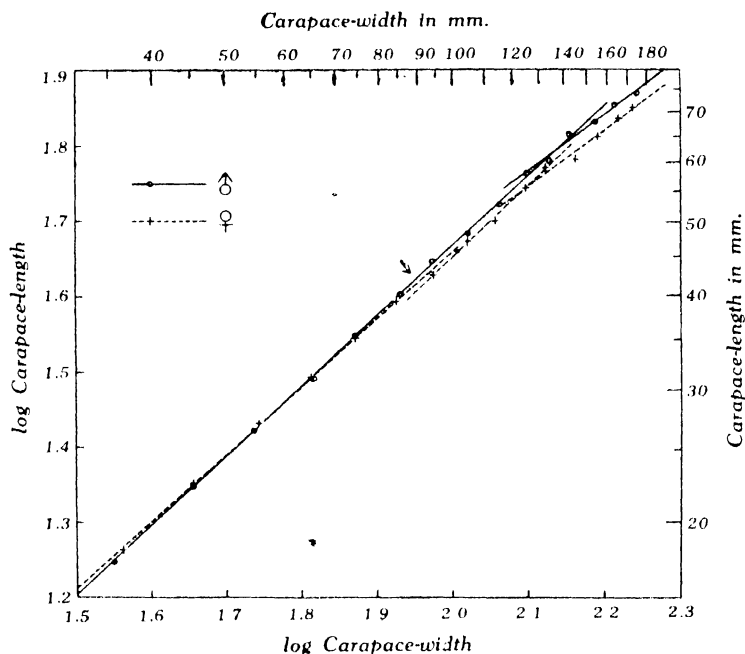


FIG. 4. The allometry of *Callinectes sapidus*. Constructed from the data of GRAY and NEWCOMBE (1938 a).

ANDERSON, LUMER and ZUPANCIC (1937) have observed that the absolute growth of *Daphnia pulex* traces an S-shaped curve in relation to instars, and MACKAY and WEYMOUTH (1935) have also observed this fact in *Cancer magister*. The allometric curve, then, necessarily breaks at several points. In fact, DAWES and HUXLEY (1934) have indicated the progressive changes of allometry from $\alpha=1.99$ to 0.47 in the crusher, and from 1.34 to 1.49 and further from this to 0.59 in the nipper in the male chelae of *Alpheus dentipes*. ANDERSON, LUMER and ZUPANCIC (1937) and WEYMOUTH and MACKAY (1934, '36) have successfully determined the relative growth by the method of applying the allometric equation together with considerations of life history or growth in relation to instars.

According to WEYMOUTH and MACKAY (1936), the log-log plot of various dimensions does not fit a straight line, but indicates a continuous curve, some parts of this curve being however approximately a straight line. Accordingly, they have mentioned that the size of the male crabs at which sexual maturity occurs is inconclusive for *Cancer magister*, on account of there being no data regarding life-history. HAMAI (1937) has shown that the relative growth ratio continuously changes with age. It

is, then, clear that a conclusion regarding critical points cannot be drawn, if the observations of growth have not been made together with those of life history or physiological facts. In the present case of *Sesarma* some observations about sexual maturity in the females and the variation in size have been made.

The abdomen of the females of *Sesarma* covers only the ventral depressed part of the cephalothorax when young, but it grows, during the younger stage, covering the ventral surface of the cephalothorax, with an intensive relative growth ratio, and it attains to almost the same size as the width of the ventral surface of the cephalothorax. This is comparable with female *Uca*, in which, while the allometry is initiated at the beginning of post-larval life, a state of equilibrium is eventually attained, when the lateral margins of the abdomen have reached the bases of the legs (HUXLEY, 1932), and it is also comparable with *Maia squinado* (Table 6). After the attainment of this size sexual maturity is reached. In fact, the smallest crab bearing eggs was 18 mm. in carapace-width. The allometry of the specimens above about 20 mm. in carapace-width, viz. that of the older stage, is, then, that of sexual maturity, and the intensity of relative growth lowers from $\alpha=1.5$ of the younger stage, to $\alpha=1.0$ (isometry) of maturity.

TEISSIER (1935) has observed in *Maia squinado* three stages of growth, of which the first stage shows $\alpha=1.31$, the second $\alpha=1.27$ and the third $\alpha=1.00$, in the abdomen width. The last mentioned ratio is that of the crabs which have attained sexual maturity, which shows "dysharmonie de taille". That is, after attainment of sexual maturity *Maia squinado* does not moult. GRAY and NEWCOMBE (1938 b) have also observed that the female blue crab, *Callinectes sapidus*, apparently does not moult after reaching maturity. In *Sesarma (Holometopus) dehaani*, however, after reaching sexual maturity one or more moultings seem to occur. WEYMOUTH and MACKAY (1934, '36) have shown that the sudden change in the length-width proportions indicates that sexual maturity is usually attained by a single moult, and that also several moultings occur after reaching maturity, in *Cancer magister*. The sudden change in the abdomen-width/carapace-width relation in *Sesarma (Holometopus) dehaani* also indicates that sexual maturity is attained by a single moult.

The present observations in the male *Sesarma* may not give a clear conclusion regarding sexual maturity because of there being no other data except growth, but the significantly great change of α in the chela-length suggests the fact that the moulting of maturity occurs at about

TABLE 6
Values of α in various species

Species	α	Reference
CARAPACE-LENGTH/WIDTH RELATION		
<i>Callinectes sapidus</i>	♂ 0.93~0.75 ♀ 0.90~0.94~0.73	Calculated from GRAY and NEWCOMBE's data (1938 a).
<i>Cancer magister</i>	both sexes (young specimens) 0.76~0.94 ♂ 0.94 ♀ 0.94 or more	WEYMOUTH and MACKAY (1936)
<i>Carcinus maenas</i>	about 1.0	HUXLEY (1932)
<i>Ocypoda aegyptiaca</i>	♂ 1.04~1.12 against anterior width ♂ 0.77~1.06 against posterior width	SANDON (1937), calculated from the reverse relations.
<i>Pachygrapsus marmoratus</i>	about 1.0	HUXLEY (1932)
<i>Sesarma (Holometopus) dehaani</i>	♂ 1.03~1.09 ♀ 1.05	Present paper
<i>Telmessus acutidens</i>	♂ 1.05 ♀ 1.19	URITA (1936), calculated from the reverse relations.
<i>Telmessus cheiragonus</i>	♂ 1.06 ♀ 1.06 ♂ 1.04 ♀ 1.05	.. SASAKI (1928), calculated from the reverse relations.
ABDOMEN-WIDTH/CARAPACE-LENGTH RELATION		
<i>Cancer magister</i>	6th segment ♂ 1.02 7th segment ♂ 1.02 ♀ ca. 1.2	WEYMOUTH and MACKAY (1936), calculated from the carapace length/width and abdominal seg./carapace width relations.
<i>Carcinus maenas</i>	unsexable and young ♀ 1.26 older ♀ 1.42 ♂ 1.07~0.94 3rd seg. ♂ 1.35~0.95 ♀ 1.33~1.08~1.29~1.08 against $\sqrt{\text{length} \times \text{width}}$	HUXLEY (1932) DAY (1936)
<i>Eriocheir japonicus</i>	5th seg. ♂ 0.90 ♀ 1.84~1.74~1.02	Calculated from the data given by OKADA and MIYASHITA (1935)
<i>Maia squinado</i>	♀ 1.34~1.27~1.00	TEISSIER (1935)
<i>Sesarma (Holometopus) dehaani</i>	3rd seg. ♂ 1.01~0.86 ♀ 1.43~0.97	Present paper, calculated from the carapace length/width and the abdomen-width/carapace-width relations.

Species	α	Reference
<i>Telmessus cheiragonus</i>	\uparrow 2nd seg. 1.10 3rd seg. 1.10 4th seg. 1.14 5th seg. 1.14 6th seg. 1.10 7th seg. 1.00 \mp 2nd seg. 1.19 3rd seg. 1.23 4th seg. 1.32 5th seg. 1.34 6th seg. 1.36 7th seg. 1.15	SASAKI (1928)
CHELA-LENGTH/CARAPACE-LENGTH RELATION		
<i>Alpheus dentipes</i>	$\sqrt{\text{propus width} \times \text{propus thickness}}$ \uparrow crusher 1.99~0.47 \uparrow nipper 1.34~1.49~0.59	DAWES and HUXLEY (1934)
<i>Cancer magister</i>	propus \uparrow 1.07~1.14 \mp 1.0	WEYMOUTH and MacKAY (1936)
<i>Ericheir japonicus</i>	\uparrow 1.62 \mp 1.25	Calculated from OKADA and MIYASHITA's data (1935)
<i>Maia squinado</i>	propodite \uparrow 1.10~1.35~1.90 \mp 1.09~1.16~1.15	TEISSIER (1935)
<i>Ocypoda aegyptiaca</i>	\uparrow large chela (ventral) 1st seg. 0.85~1.31 2nd seg. 1.20~1.46 3rd seg. 1.21~1.23 4th seg. 1.03~0.89 5th seg. 1.40~1.2 6th seg. 1.04~1.05 7th seg. 1.08~1.05 \uparrow small chela (ventral) 1st seg. 1.05~1.15 2nd seg. 0.91~1.33 3rd seg. 0.96~1.18 4th seg. 0.86~0.90 5th seg. 1.31~1.29 6th seg. 1.00~0.99 7th seg. 0.97~0.93	SANDON (1937)
<i>Sesarma (Holometopus) dehaani</i>	propodite \uparrow 1.17~1.33 \mp 1.10	Present paper, calculated from the carapace length/width and chela-length/carapace-width relations.
<i>Telmessus cheiragonus</i>	propodite right \uparrow 0.98 \mp 0.97 left \uparrow 1.00 \mp 0.98	SASAKI (1928)
<i>Upogebia littoralis</i>	\uparrow 1.37~1.13 \mp 1.05~0.90	HUXLEY (1932)

16 mm. or more in carapace-width. Regarding the relative growth of the chela, TEISSIER (1935) has given the values of α against the carapace-length as follows: in *Maia squinado*, at the first stage, $\alpha=1.10$, at the second stage, $\alpha=1.35$, and at the third stage, $\alpha=1.90$. In *Cancer magister*, in the first stage, $\alpha=1.07$, and in the second stage, $\alpha=1.14$, against the carapace-width (WEYMOUTH and MACKEY, 1936); in *Ocypoda aegyptiaca*, in the large chela, α of the first segment is $1.10\sim1.54$, that of the fourth segment is $1.05\sim1.24$, and in the small chela, the first segment has $\alpha=1.06\sim1.40$, the second segment $0.99\sim1.61$, the third segment $1.00\sim1.68$, the fourth $0.89\sim1.16$, and the sixth $0.99\sim1.16$ against the anterior width of carapace. Values of α above mentioned all indicate the rise, viz. the changes increasing the relative growth intensity from the younger stage to the older, as seen in the present case. High positive allometry and rise of α in male chela would be a general tendency when reaching maturity (Table 6).

The high growth intensity is maintained by the chela in comparison with the slight allometry or isometry of the abdomen in male *Sesarma* throughout the younger and older stages. In the females, while the chela indicates a weak positive allometry throughout all the stages, the abdomen grows very rapidly during the immature stage. As the result of these growth features, the female crabs obtain harmonious proportions of various parts at the older stage of their existence, while the males gradually lose harmonious proportions as they grow older (Table 2 and 3). GRAY and NEWCOMBE (1938 a) consider that the carapace-length constitutes a better standard-index of size than the chela-length, the eye-to-spine-width or the carapace-width in *Callinectes sapidus*. In *Sesarma (Holometopus) dehaani*, the carapace-length and the carapace-width seem to be of a similar value as the standard-index of size and of growth.

SUMMARY

1) The four dimensions, viz. the carapace-width, the carapace-length, the width of the third abdominal segment, and the propus-length of the right chela were measured; and the carapace-width having taken as the standard, the relative growth of each part has been discussed.

2) Two stages of growth are distinguishable, with the critical points at about 16 mm. carapace-width in the males, and at about 20 mm. carapace-width in the females.

3) The abdomen-width in female crabs shows a high positive allometry

in the younger stage, but, entering the older stage, becomes parallel with the slight allometry or isometry of the other dimensions.

4) The female crabs attain sexual maturity at the size of about 20 mm. in carapace-width.

5) The male chela intensifies its positive allometry on entering the older stage, and becomes finally of the same size or more as the carapace-length. On the other hand, the carapace-length and the abdomen-width in the males are slightly allometric or isometric.

6) The analytical method of relative growth by means of the allometric equation has been discussed, where the dimensional allometry is in question.

REFERENCES

- ANDERSON, B. G., H. LUMER and I. J. ZUPANCIC, JR. 1937. Growth and Variability in *Daphnia pulex*. Biol. Bull., Vol. 73, pp. 141-163.
- DAWES, B. and J. S. HUXLEY. 1934. Rapid Growth-Rate and Diminishing Heterogony. Nature, Vol. 133, p. 982.
- DAY, J. H. 1936. Growth Gradients in the Abdomen of the Shore-Crab. Nature, Vol. 137, p. 688.
- GRAY, E. H. and C. L. NEWCOMBE. 1938a. The Relative Growth of Parts in the Blue Crab *Callinectes sapidus* RATHBUN. Growth, Vol. 2, pp. 235-246.
- GRAY, E. H. and C. L. NEWCOMBE. 1938b. Studies of Moulting in *Callinectes sapidus* RATHBUN. Growth, Vol. 2, pp. 285-296.
- HAMAI, I. 1937. Some Notes on Relative Growth, with Special Reference to the Growth of Limpets. Sci. Rep. Tôhoku Imp. Univ. Biol., Vol. 12, pp. 71-95.
- HAMAI, I. 1938. Relative Growth of *Meretrix meretrix* and its Local Variation, as shown by Experiments in Rearing. Sci. Rep. Tôhoku Imp. Univ. Biol., Vol. 13, pp. 205-220.
- HAMAI, I. 1939. On the Growth of the Embryonic Shell of the Japanese River Snail, *Viviparus japonicus*. Sci. Rep. Tôhoku Imp. Univ. Biol., Vol. 14, pp. 1-10.
- HUXLEY, J. S. 1932. Problems of Relative Growth. London.
- HUXLEY, J. S. and G. TEISSIER. 1936. Terminologie et Notation dans la Description de la Croissance Relative. Com. Rend. Soc. Biol., Tome 121, p. 934.
- MACKAY, D. C. G. and F. W. WILMOUTH. 1935. The Growth of the Pacific Edible Crab, *Cancer magister* DANA. J. Biol. Bd. Can., Vol. 1, p. 191-212.
- NEEDHAM, A. E. 1937. On Relative Growth in *Asellus aquaticus*. Proc. Zool. Soc. London, Ser. A, 1937, pp. 289-313.
- OKADA, YÔ K. and Y. MIYASHITA. 1935. Succulnization in *Eriocheir japonicus* DE HAAN, with Remarks on the Occurrence of Complete Sex-reversal in Parasitized Male Crab. Mem. Coll. Sci. Kyôto Imp. Univ. Ser. B, Vol. 10, pp. 169-208.
- PAULIAN, R. 1936. L'Existence d'un Stade Critique dans la Croissance Relative de l'*Eupagurus prtdauxii* (Crustacé Anomoure). Com. Rend. Soc. Biol., Tome 121, pp. 435-437.
- SANDON, H. 1937. Differential Growth in the Crab *Ocypoda*. Proc. Zool. Soc. London, Ser. A, 1937, pp. 397-414.

- SASAKI, K. 1928. On the Growth of *Telmessus cheiragonus* (TILSIUS). Sci. Rep. Tōhoku Imp. Univ. Biol., Vol. 3, pp. 805-819.
- TEISSIER, G. 1935. Croissance des Variants Sexuels chez *Maia squinado* L. Trav. Stat. Biol. Roscoff., Fasc. 13, pp. 93-130.
- URITA, T. 1936. Dimensional, Morphological and Zoogeographical Study of Japanese Crabs of the Genus *Telmessus*. Sci. Rep. Tōhoku Imp. Univ. Biol., Vol. 11, pp. 69-89.
- WEYMOUTH, F. W. and D. C. G. MACKEY. 1934. Relative Growth in the Pacific Edible Crab, *Cancer magister*. Proc. Soc. Exp. Biol. Med., Vol. 31, pp. 1137-1139.
- WEYMOUTH, F. W. and D. C. G. MACKEY. 1936. Analysis of the Relative Growth of the Pacific Edible Crab, *Cancer magister*. Proc. Zool. Soc. London, 1936, pp. 257-280.

BRIEF OBSERVATIONS ON THE RAPID MULTIPLICATION OF *CYCLIDIUM* SP. IN PUTREFYING SEA-WATER OF KNOWN PH AND SALINITY¹

By

ZENZO MATSUYA

The Marine Biological Station, Asamushi, Japan

With 2 Text-figures

INTRODUCTION

In the study of plankton one may often meet with the fact that minute protozoan organisms, especially *Cyclidium* sp. belonging to Pleuronemidae of Ciliata, appear in abundance in samples of sea-water which have been kept for a few days (Fig. 1). *Cyclidium* may hardly be observable under the microscope when the sample is fresh, but with the lapse

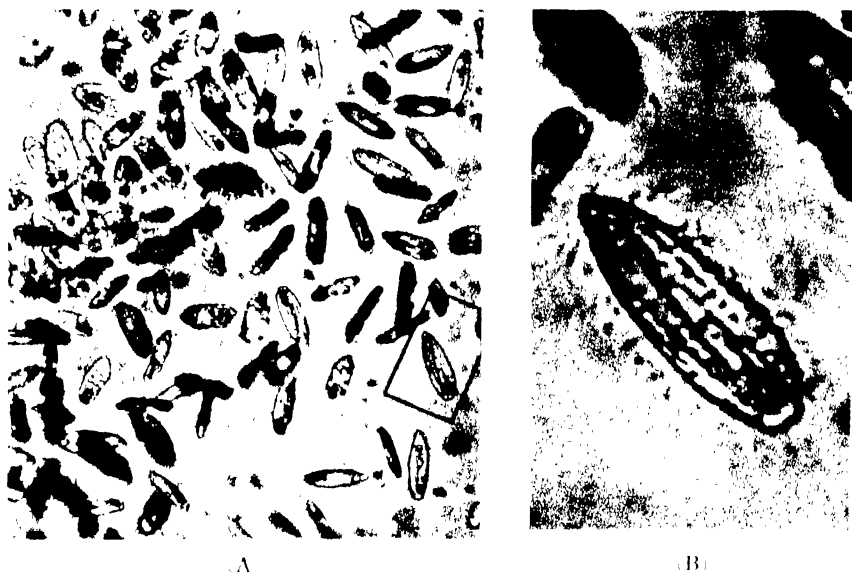


Fig. 1. *Cyclidium* sp. (Specimen killed by heating gently). A) ... showing multiplication (ca. $\times 290$). B) ... showing a portion of A), enlarged about four times.

Photo. Dr. S. KOKURO

¹ Contribution from the Marine Biological Station, Asamushi, Aomori-ken, No. 171

of time it appears in great abundance, as the decaying plankton affords favorable culture-media for this organism. Such rapid multiplication seems to be usual in some protozoan species, and in most cases this phenomenon is accompanied by chemical changes in the water such as decrease of the pH and dissolved oxygen, these changes due probably to the decomposition of organic matter caused by bacteria. Besides bacteria, however, saprozoic forms or bacterial feeders subsequently appear and, finally, dominate the Diatoms in the container, suggesting the production of nitrite and nitrate from organic nitrogen compounds.

In the present article the appearance of *Cyclidium* sp. in putrefying sea-water was particularly studied as a problem. NOLAND (1925), MORGAN (1926), FINLEY (1930), LACKEY (1936, 1938), OKADA and other (1939), and UEMURA (1939) studied the ecology of *Cyclidium glaucoma*, from different habitats. From this fact it can be noted that *Cyclidium* is a resistant form, capable of adjusting itself to and tolerating environmental changes to some extent.

The present brief observations were carried out with the object of studying the range of changes in the pH and salinity of the medium in which *Cyclidium* lives and multiplies.

Taking this opportunity I wish to express my hearty thanks to Dr. S. KOKUBO who suggested the problem and directed the work. Thanks are also due to Prof. S. HOZAWA, Director of our station, under whose supervision the present investigation was made.

MATERIAL AND METHODS

After thoroughly mixing a large quantity of fresh plankton with a certain amount of sea-water, a definite volume of it was poured in several flasks (Erlenmeyer, 300 c.c. in capacity) a set of which, after being adjusted to a desired pH and salinity according to the following procedures, was kept in the laboratory. Within a few days, *Cyclidium* appeared in the media of suitable pH and salinity, accompanied by an increase of bacteria, which caused the upper layer of media to become gradually opaque. To ascertain the appearance of *Cyclidium*, a few drops of sea-water, taken from the surface and from the bottom of the flask once a day, were observed qualitatively under the microscope. Thus, the ranges of the pH and salinity of medium where the multiplication of *Cyclidium* took place were determined, and also, the time required for the appearance of *Cyclidium* and the duration of its survival were noted. During the

observation the changes in the pH of the medium were recorded by means of the indicator method.

To regulate the pH of sea-water, buffer-solution with an adequate pH was added drop by drop to the 10 c.c. of sea-water until the latter showed a desired pH. From the amount of buffer-solution needed in doing this, the pH of any large quantity of sea-water could be regulated by calculation. Observations were made at each pH value of 2.2, 2.6, 3.0, 3.6, 4.0, 4.6, 5.0, 5.8, 6.6, 7.0, 7.8, 8.2, 8.8, and 9.4. These pH values were regulated by addition of one of the following buffer-solutions to sea-water: — citrate-HCl (pH 1.2, 2.0, 3.0, 4.0, 5.0 and 6.0), phosphate-phosphate (pH 7.0), borate-HCl (pH 8.0) and borate-NaOH (pH 9.0 and 10.0). For the alkaline-range with the pH higher than 9.4 (Exp. 2) the regulation was made by adding NaOH solution of pH ca. 11.0, which was made by mixing conc. NaOH with sea-water and removing the resulting precipitate of magnesium hydroxide. As the pH of sea-water thus regulated tended to change with time, a proper amount of buffer-solution was added once a day to maintain the initial pH.

In the experiments relating to the salinity, distilled water, condensed sea-water and the hypertonic saline solution of vant't Hoff were used to obtain various concentrations of sea-water. To prepare sea-water of low salinity, natural sea-water was diluted with distilled water, while sea-water with high salinity was prepared by adding an adequate quantity of condensed sea-water or van't Hoff's solution to natural sea-water. The salinity of the sea-water used was accurately calculated from the chlorinity determined by MOHL's method. Thus, experiments were carried out with sea-water of various salinity, ranging from 1.3 to 90.3‰.

Concerning the development of *Cyclidium* in relation to its heat-resistance, only one experiment was carried out. In this case the flask containing plankton was kept for one hour in the thermostat at a definite temperature. Then, the flask was taken out and placed in the laboratory at room-temperature, and the limit of high temperature which *Cyclidium* could not survive, was roughly determined. The temperatures examined were 29°, 32.5°, 35°, 38.5°, 40°, 45°, 51.5°, 58°, and 69°C.

The present observations were made during the period of four months from April to July, 1938. In the course of the experiments, the room-temperature showed marked fluctuations (Max. 29.2°–Min. 13.4°C.), and plankton material used as the source of organisms in the flask also varied in quantity as well as in species. But differences in these factors seemed to have no great effect on the appearance and growth of *Cyclidium*.

EXPERIMENTAL RESULTS

Observation 1. Appearance of *Cyclidium* related to the pH of sea-water.

As is shown in Table 1, the ranges of pH, in which *Cyclidium* could live and multiply, were from pH 5.8 to 9.8, but it extended from initial pH 5.0 to 9.8, when daily changes in the pH were admitted. In those ranges *Cyclidium* was found after 2 to 4 days, and it multiplied rapidly and could live actively for a long period. The duration of survival varied with the pH, generally lasting longer than 2 weeks.

TABLE 1

Initial pH	Experiment 1				Experiment 2			
	Days elapsed before appearance	pH after one day	pH after five days	Duration of survival (in days)	Days elapsed before appearance	pH after one day	pH after five days	Duration of survival (in days)
2.2		2.2	2.2	—	*	*	*	*
2.6		2.6	2.6	—	*	*	*	*
3.0		2.8	3.0	—	*	*	*	*
3.6		3.3	3.7	—	*	*	*	*
4.0		3.6	4.0	—	*	*	*	*
4.6		4.2	4.7	—	*	*	*	*
5.0		4.8	5.2	—	*	*	*	*
5.8	1	5.9 ₅	5.8	13	*	*	*	*
6.6	3	6.5	6.6 ₅	14	*	*	*	*
7.0	2	7.2	7.0 ₅	15	*	*	*	*
7.8	4	7.6 ₅	7.7	13	*	*	*	*
8.2	3	7.7	8.1	16	2	6.9	7.7 ₅	9
8.6	*	*	*	*	2	6.8	8.4	19
8.8	1	8.7	8.7 ₅	17	*	*	*	*
9.0	*	*	*	—	2	6.7	8.7 ₅	15
9.1	1	9.1	9.3	17	2	6.9	9.2	17
9.8	*	*	*	*	4	7.3	9.3	15
10.2	*	*	*	*	—	7.7 ₅	9.2	—
10.4	*	*	*	*	—	8.4	9.4	—
10.8	*	*	*	—	—	9.3	9.8	—
Control	2	7.4	—	19	3	6.4	7.3	19

The pH and the mode of appearance of Cyclidium. The result will be described only of representative experiments (Table 1). In Experiment 1, it was found that within one day after the experiment was started the plankton settled down on the bottom of the flask, its colour being brown at first, but later, on account of partial decolorisation, becoming yellowish-white in acid media, and yellowish-brown in neutral and alkaline media, the colour-tint increasing with the pH of the media. After one or two days, the water became gradually turbid, and micro-organisms appeared

in the flasks with the initial pH of 5.8-8.8, but no *Cyclidium* as yet appearing. *Cyclidium* was first found in the flasks of pH 7.0 and also in the control-flask at the end of two days. On the next day, the flasks of pH 6.6 and 8.2, and after four days all the flasks (pH 5.8 to 9.1) showed the presence of *Cyclidium*, which immediately increased rapidly in number. In the flasks of pH 5.0 and below, no *Cyclidium* was found during the long period of observation, although bacteria developed in these low values of pH. The lowest limit for the appearance of *Cyclidium* was pH 5.8 in Experiment 1, but in another experiment, in which daily regulation of pH was not made, it was 5.0, showing the tendency that, with the lapse of time, the media with initial pH of 6.8 and below showed an increase of pH, while those with initial pH 7.8 and above showed a rapid decrease of pH. With such rapid appearance of *Cyclidium*, the turbidity and odour of the sea-water increased, especially emitting the odour of H_2S from the flasks of pH 7.8, 8.2 and from the control-flask in which the plankton became greenish black. Thus *Cyclidium* multiplied rapidly for a few days, but soon tended to become gradually small in size, less active and decreasing in number, at last resulting in extinction. The duration of the survival of *Cyclidium* in the flasks was generally about two weeks or more, though it varied slightly with pH, and from this time onwards the water became clear and the odour vanished owing to the self-purification action of sea-water.

In Experiment 2, where the desired pH of sea-water was made by adding NaOH-sea-water solution, *Cyclidium* was found in the flasks of pH 8.2-9.4 after two days, and in the flask of pH 9.8 after four days. Moreover, the duration of survival was longer than two weeks as in the results of Experiment 1. In Experiment 2, it is noteworthy that the pH in all flasks showed remarkable changes, which had never been seen in Experiment 1. For example, the pH 9.8, limit for appearance of the organism in the alkaline range, fell to pH 7.3 and even so high a pH as 10.2, in which no *Cyclidium* were found, fell rapidly to pH 7.7, after one day, and it further decreased with the lapse of time. This was caused probably by the acidity due to CO_2 , which was produced by micro-organisms.

pH-behavior. As was already found in Exp. 1 and 2, the change of pH caused by the metabolism of living organisms was unavoidable even in the sea-water whose pH was daily regulated to maintain the initial pH. When daily regulation was not made, the pH-change of natural sea-water was very regular. Table 2 and Figure 2 show the results of experiments

for a period of 25 days, which were carried out in order to observe the pH change of the medium in flasks (A), (B), (C) and (D), containing 93%, 46%, 23% and 5% volumes of plankton respectively.

TABLE 2

Experiment 4

Time (in days)	Volume of plankton							
	93% (A)		46% (B)		23% (C)		5% (D)	
	pH	Cycl.	pH	Cycl.	pH	Cycl.	pH	Cycl.
1	6.4 ₅	—	6.5	—	6.8 ₄	—	8.1 ₅	—
2	6.4	+	6.5	+	6.9	+	7.7	—
3	6.7	+	7.0 ₅	+	7.1 ₅	+	7.6 ₅	+
6	7.2	+	7.3	+	7.4 ₅	+	7.7	+
8	7.3 ₅	+	7.3 ₅	+	7.5	+	7.9	—
14	7.7	+	7.8 ₅	+	7.9 ₅	+	9.2	—
18	7.8 ₅	+	8.9	+	9.5	—	9.2	—
20	8.0	+	9.5	+	9.2 ₅	—	8.9	—
25	9.4	+	8.9	—	8.8	—	8.4 ₅	—

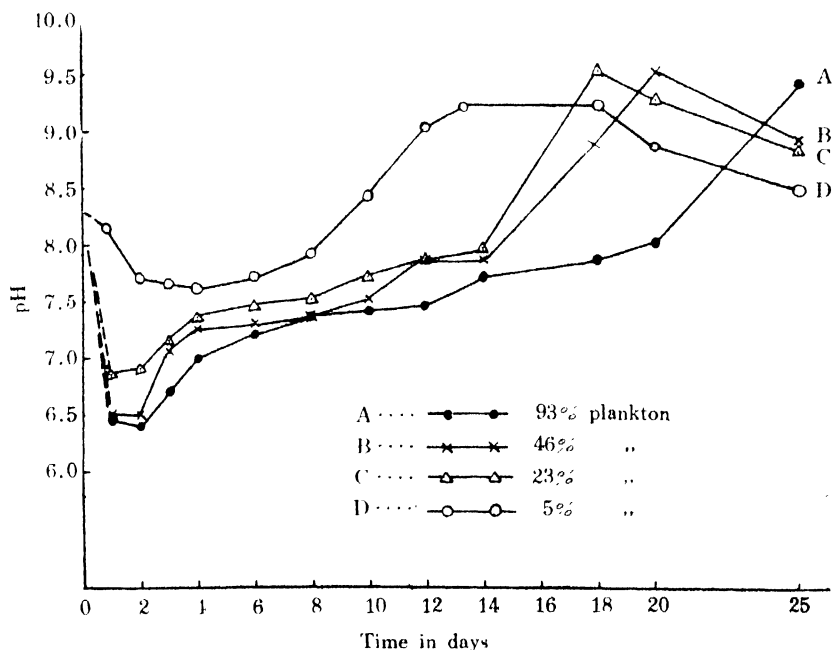


Fig. 2. Curves showing the relation between the quantity of plankton and the change of pH.

Looking at the table and the figure, one will note that the course of the pH change in sea-water was almost similar in each case, and could be divided into the following four stages.

In the first stage, the acidity of the water rose, *i. e.* the pH rapidly fell to the lowest value. Though this pH varied according to the quantity of plankton present it was 6.4₅ in A, 6.5 in B, 6.8₅ in C and 7.6₅ in D. The lowest pH was attained within two days except in D. Bacteria but no *Cyclidium* was found in this stage.

Following the lowest pH or turning point, the second stage began. In this stage, the pH rapidly rose to neutrality and then gradually became alkaline. The rate of increase in pH and the period of this stage varied according to the quantity of plankton present. *Cyclidium*, as well as other organisms, began to appear at the end of the first stage and at the beginning of the second stage, and at the same time, the water became turbid and emitted a putrescent odour. *Cyclidium* and bacteria, which increased enormously in number and were very active from the beginning to the middle of this stage, tended to decline from the end of this stage.

In the third stage, where the multiplication of diatoms such as *Navicula* and *Nitzschia* took place, the pH rose rapidly to the maximum, probably due to the decrease of CO₂ owing to its consumption in the photosynthesis by diatoms. The maximum pH was observed to be 9.4 on the 25th in A, 9.5 on the 20th in B, and on the 18th in C, and 9.2 on the 14th day in D respectively after the experiment was started. During the period of this stage, the putrescent odour gradually diminished and the water became clear. *Cyclidium* was absent or scarcely present.

In the fourth stage, the pH again fell rapidly. *Cyclidium* was still found in the sea-water containing a large amount of plankton as in A, but it completely disappeared in B, C and D. The water became quite clear, evolving no odour at all.

The four stages of the pH-change above mentioned were always observable in these experiments, although the length of the period of each stage varied in each case, and the boundaries of the stages were indistinct in the sea-water which contained but little plankton as in D. Thus it was notable that the quantity of plankton, which had been inoculated and which had developed the micro-organisms, influenced the rate and duration of each stage.

Regarding the pH change, PRUTHI and JONES obtained almost similar results. PRUTHI (1927), who studied the relation between the pH of

hay-infusion and the protozoan population without seeding, reported that the rate of change in the pH of the infusion depended upon the nature of the water. JONES (1930) studied the infusion histories of *Paramecium* from the view point of hydrogen ion changes and states that "... This acid phase is accompanied by fermentation, and is evidently brought about by bacterial activity.... The second stage of the pH behavior, in which the infusion returns to normality and then becomes alkaline, is probably brought about by a second cycle of bacteria." Moreover, the prolonged duration of the acid condition in the medium, has an important bearing upon the fate of organisms, as was stated by JONES. In the above experiments already described, the acid limitation for the appearance of *Cyclidium* varied slightly, depending on whether or not a daily regulation of pH was made, that is, no *Cyclidium* was found in sea-water of initial pH 5.0 when daily regulation pH was applied. But the appearance of *Cyclidium* was observed in sea-water of the same pH when no daily regulation was made. This fact will be understood from the reasons above mentioned.

Relation between vital phenomena of the other ciliates and pH ranges. The above experiments show that *Cyclidium* can live and multiply for a long period in wide ranges of pH of the medium. Such a phenomenon was observed also in other ciliates, although it differs with the species and the conditions of experiments.

From the ecological standpoints, NOLAND (1925), who studied the distribution of 65 species of fresh-water ciliates, observed that some of them have great tolerance for variations in temperature, in oxygen content, and in hydrogen ion concentration of the environment. According to him, *Cyclidium glaucoma* was found to inhabit the range of pH 6.5-8.6. According to LACKY (1938), who studied some factors affecting the distribution of the protozoan, *Cyclidium glaucoma* was found in varieties of habitats such as ponds, pools, oceans, polluted streams and so on, where the pH ranged from 2.9 to 8.0.

On the other hand, according to many investigators the relation of the vitality of the ciliates to the pH of experimental medium was found to be as follows:--

<i>Spirostomum ambiguum</i> (Growth)	pH 6.8-7.8....	SAUNDER '24
" " "	pH 6.5-8.0	MOREA '27
<i>Colpidium striatum</i>	pH 4.0-8.6 ...	ELLIOTT '33
<i>Colpidium</i> sp.	pH 6.0-8.5.....	PRUTHI '27

<i>Paramaecium aurelia</i>	(Growth)	pH 5.7-7.8.....	DARBY	'29
"	(Living)	pH 6.0-9.5.....	MOREA	'27
<i>P. multimicronucleatum</i>	"	pH 4.8-8.3.....	JONES	'30
<i>P. caudatum</i>	(Division)	pH 5.3-8.2.....	DARBY	'29

Looking through the above table, we know that the vital phenomena of the ciliates take place in wide ranges of pH, although the differences in the pH range were observed in different species and in different phenomena, and that, in the present experiment, the ranges of pH in the sea-water in which *Cyclidium* multiply and live were practically similar to those in MOREA's observation made on *Paramecium aurelia*. Moreover, it is noteworthy that a single or double maxima in the pH for these phenomena were observed. ELLIOTT (1933), studying the growth of *Colpidium striatum* in relation to the pH of the medium, observed two growth maxima within the pH range 4.0 to 8.6, one above and one below pH 7.0, and they were replaced by a single maximum if sodium acetate was added to the media. The presence of two optima in the pH was also reported by MAST (1931) on the longevity of *Amoeba proteus*, one being pH 6.6-7.0, and the other pH 5.0-6.4. Thus, as the viability and the rate of growth varied with the pH of the medium, we may suppose that one or two optima pH for *Cyclidium* may exist, though, in so far as the results of the present investigation are concerned, this point has not yet been determined.

Observation II. Relation to salinity.

As the results of two experiments showed a strong coincidence, they will be represented by Experiment 6, which is given in Table 3.

The range of salinity as well as the range of pH in which *Cyclidium* can normally live, is very wide, the former being as wide as 7.8 to 54.4‰.

In succession to the bacterial development, *Cyclidium*, which first appeared around the natural salinity, was found in the media of 22.9 to 37.9‰ salinity after two days, but this range became wider, ranging from 7.4 to 54.4‰ on the fourth day. But no *Cyclidium* was found in the media of salinity below 7.4‰ and above 74.7‰ during the period of a 25-day observation. In the flasks of 16.1‰ salinity and above, *Cyclidium* could live actively for more than 13 days on an average, while in flasks of 7.4‰ and 10.7‰ it lived only for one day and then disappeared. In Experiment 5, however, where *Cyclidium* was found in all the ranges

TABLE 3

Experiment 6

Salinity (‰)	Days elapsed before appearance	pH after one day	pH after five days	Duration of survival (in days)
1.3	-	6.6 ₁	6.4	-
6.7	-	6.1 ₅	6.7	-
7.4	3	6.1 ₅	6.7	1
(7.8)	(3)	(7.4)	(6.7)	(11)
10.7	3	6.3	6.5 ₅	1
(10.7)	(4)	(7.0 ₅)	(6.8 ₅)	(10)
16.1	3	6.1 ₅	6.9	12
(16.1)	(4)	(7.4 ₁)	(6.9)	(15)
22.9	2	6.7 ₅	6.5	22
30.2	2	6.2	7.1 ₅	15
31.8	2	6.4 ₅	7.2	15
37.9	2	6.2	7.0	17
53.8	3	6.2	6.9	12
54.4	3	6.7	6.7 ₅	23
74.7	-	7.1	7.0	-
78.2	-	6.9 ₅	6.6 ₅	-
90.3	-	7.5	6.8	-

Figures in the brackets show a part of the result obtained from Exp. 5

examined, and a little later than in Exp. 6, the survival of *Cyclidium* continued for 14 days on an average, and for about 10 days even in 7.8‰ and 10.7‰ salinities. Such a difference of duration in the medium of 10.7‰ salinity may perhaps be due to the differences in temperature and possibly to the plankton contents of the media. At any rate, the lowest limit for the appearance was the salinity of 7.8‰, corresponding approximately to a 4-fold diluted sea-water. On the other hand, *Cyclidium* was very resistant to higher salinities as well as to lower salinities, and lived in the salinity of 54.4‰ for 23 days or longer, but in the salinity of 74.7‰ the multiplication was completely checked. Namely, the limitation for high salinity seems to correspond to about a 2-fold concentrated sea-water. Moreover, there was no marked difference in the duration of the survival whether the medium was prepared from the condensed sea-water or from a mixture of sea-water and van't Hoff's solution.

The pH change in the medium was on the whole similar to the results of Exp. 4, showing the four stages described in that experiment. The lowest pH in the first stage found, after a few days, varied from 6.1 to 6.4 in Exp. 6, and from 6.6₅ to 7.6₅ in Exp. 5. The highest pH observed, after the disappearance of *Cyclidium*, differed also with the salinity, being generally higher than 8.6. In the media of extremely

high or low salinity, where *Cyclidium* was not found, the changes in the pH were very gradual, never showing four stages, and the pH generally being below 8.0.

The rate and duration of diatom growth and their relation to pH change were on the whole similar to those observed in Exp. 2 and Exp. 4.

Thus, *Cyclidium* could well adapt itself to the salinity changes caused by the artificial dilution or condensation of sea-water, suggesting the strong viability of this organism in varying salinities in natural environment. This may probable be due to its simple organization, which may allow it to adjust itself to outer conditions with much ease.

Concerning the resistance of the organisms in relation to the varying salinity of the media many investigations have been made by scientists with regard to lower aquatic animals. ANDREWS (1925) observed that the resistance of marine animals such as the crab, the limpet and the green sea-urchin to fresh-water differed in species and ages, and even in the same species, the smaller the animal, the shorter was the survival time. PEARSE (1928), studying the viability of *Limulus*, *Phascolosoma* and eighteen marine annelids in diluted sea-water, obtained this result: that most of them lived for one or two weeks in a mixture of three-fourths sea-water and one-fourth freshwater, but that some of them viz: *Nereis virens*, *Limulus polyphemus* and *Laonice viridis* lived for the period of two or three weeks in one-fourth sea-water and three-fourths fresh-water. HIRO (1938) reported recently that the resistance of some littoral barnacles to altered salinity differed with species and habitat. On protozoan species, WORLEY (1929) observed that the marine rotifer, *Brachionus murei* collected from an alkaline pond with a salinity of 4.5‰, were able to live in other alkaline ponds varying in salinity from 9.5 to about .057‰ of NaCl. According to FINLEY (1930), who studied 50 species of fresh-water protozoan with regard to their toleration to salinity, *Uronema marina*, *Colpoda aspera*, *Bodo uncinatus* and *Pleuromona jaculans* have the power to survive direct transfer into sea-water. Most of the other protozoans tolerate only a low salinity when directly transferred, but can adapt themselves, when transferred gradually, to so high a salinity as might have been fatal to them if the transfer had been direct. MAST (1931) observed *Amoeba proteus* in relation to the purity of the water, with the result that it lived for several days in water of very high purity, but survived longer in the proper concentration of salt, whether single or mixed, living to a maximum of 18 days in the former and 22 days in the latter.

Observation III. Relation to temperature.

In relation to temperature, only one experiment was made in order to study the thermal resistance of *Cyclidium*. As is shown in Table 4, the result of Experiment 7 demonstrates that the highest temperature, in which *Cyclidium* could survive an exposure of one hour, and continue normal life for a long time afterwards, was about 35°C.

TABLE 4

Experiment 7

Temperature °C.	Days elapsed before appearance	pH after one day	pH after five days	Duration of survival (in days)
Control	1	8.3	7.8	7
29°	1	8.1	7.6	7
32.5°	2	8.2	7.5 _s	12
35°	2	8.0	7.7	12
38.5°	3	7.8	7.7 _s	1
40°	-	8.0	7.7 _s	-
45°	-	8.2	7.6	-
51.5°	-	8.1	7.8	-
58°	-	8.5	7.7 _h	-
69°	-	8.7	7.7 _s	-

Within one day of the beginning of the experiment, the first few individuals of *Cyclidium* were found in the flasks of the control (21°C.) and of 29°C., increasing rapidly in number by the third day, and on the same day they also appeared in the flasks of 32.5°C. and 35°C. The duration of the survival of those individuals was seven to twelve days or more, being comparatively shorter than the periods in the preceding experiments. On the other hand *Cyclidium*, which was found in the flask of 38.5°C. after three days, disappeared soon, and had only one day's existence. At 40°C. and above, its multiplication was completely checked owing to the heat-coagulation of the protoplasm of *Cyclidium*. Regarding the pH changes, the four stages already mentioned in the preceding experiments were not distinct in this experiment. The fall in pH in the first stage was comparatively gradual while the rises in pH in the second and third stages were rapid, namely, the changes were almost similar to that of D in Exp. 4. The increase of diatoms with the rises of pH was observed after about seven days, and the maximum pH which was generally attained after 13 days was 8.8 or higher. Moreover, in this experiment some *Cyclidium* were still found in the media which showed the maxima of pH. The shortening of the duration and the rapid increase in diatoms

are certainly due to influences of slight rises of temperature under experimental conditions, and of the insufficient quantities of plankton used.

From the above experiment, one will note that when the culture was exposed to the temperature of 35°C. for one hour, no harm to the multiplication of *Cyclidium* was caused, but its survival was impossible after exposures to 40°C. and above.

As is well known, within the physiological limits, the rises in temperature of the outer medium, accelerate the vital activity of organisms, and beyond certain limitations, animals as well as organs have their own lethal or maximum temperature. Beyond this all vital phenomena cease. This maximum temperature varies with the time of exposure, namely time-factor plays an important rôle in the effect of temperature. Therefore, in the present experiment, the multiplication of *Cyclidium* would have been checked at a lower temperature than 40°C. provided the time of exposure had been prolonged.

Regarding the relation between the time of exposure and the vital phenomena at various temperatures, PORT (1927), made studies on the action of natural salts on the coagulation of protoplasm in *Paramecium caudatum*, and obtained the result that the time required to coagulate the protoplasm at the constant temperatures of 36°, 38°, and 40°C. was 2820, 1365, and 264 seconds, respectively. POYARKOFF (1928) observed that the time required for the disintegration of *Paramecium* at 36°, 38°, and 40°C. was 37.6, 8.86, and 1.81 minutes, respectively. Assuming that the checking of the multiplication of *Cyclidium* might be due to the coagulation of protoplasm at 40°C., the duration of lethal exposure for one hour or less in *Cyclidium* seems to be too long as compared with that in *Paramecium*. Namely it suggests that the heat-resistance of *Cyclidium* is markedly higher than that of *Paramecium*. In the present experiment, however, it needed a short lapse of time before the temperature reached 40°C., and consequently the time relation of temperature-effect was somewhat inaccurate in comparison with the results of the experiments of others. It is also well known that the heat-resistance or coagulation of protoplasm is affected by the pH, by the osmotic pressure, and by the nature and concentration of the salts of the medium. Hence the comparison just mentioned should not be taken as strictly accurate.

Lastly, to know the relation between the temperature-maximum and the optima of various vital activities in *Paramecium*, a few examples may be quoted from the tables in the "Temperature and living matter" (p. 165-167) of BÉLEHRÁDEK (1935), as follows:

Species	Temperature maximum	Duration of exposure	Remark	Authority
<i>P. aurelia</i>	31.5°	permanent	development	WOODRUFF and BAITSELL '11
<i>P. caudatum</i>	34-37	several hours	ciliary movement	MICHELSON '28
<i>P. caudatum</i>	33	1-2 days	"	"
<i>P. sp</i>	43	25 seconds	killing	POYARKOFF '28
<i>P. caudatum</i>	44	9 seconds	"	PORT '27

SUMMARY

1) *Cyclidium* sp. (Fig. 1), a saprozoic protozoan, is hardly observable in fresh collections of plankton, but it appears in great abundance when a large amount of plankton sample is left in putrefaction in the laboratory. In the present experiment, the development of this organism in this respect was experimentally studied, stress being laid on the pH and salinity of the medium.

2) Under ordinary laboratory conditions *Cyclidium* multiplies rapidly in sea-water, provided a rich supply of decaying plankton is available in it, and lives normally for two or three weeks.

Duration of survival depends upon the quantity of available decaying plankton. The larger the amount of plankton, the longer the duration of the survival of *Cyclidium*.

3) The range of pH in which *Cyclidium* could live and multiply for a long period was found to be from pH 5.0 to 9.8.

4) The pH of water in which *Cyclidium* is propagating shows considerable changes. The rate of change in this case is mainly due to bacterial respiration and diatom assimilation, but it also depends on the quantity of plankton in the flask.

5) The change of the pH in medium water generally shows four stages, *i. e.*, first the pH falls rapidly, secondly it gradually rises, thirdly shows a rapid rise, and fourthly again shows a rapid fall. The length of each period varies with the quantity of plankton.

6) The total salinity in which *Cyclidium* continues normal life ranges from 7.8‰ to 54.4‰.

7) One hour's exposure of the sample to a constant temperature of 35°C. does not completely depress the multiplication of this organism, but above this temperature its multiplication is depressed or checked.

LITERATURE

- ANDREWS, F. B. 1925. The Resistance of Marine Animals of Different Ages. Publ. Puget Sound Biol. St. Vol. 3, p. 361.
- BELEHRÁDEK, J. 1935. Temperature and Living Matter. Protoplasma-Monographien, Vol. 8, p. 165.
- DARBY, HUGH, H. 1929. The Effect of Hydrogen-ion Concentration on the Sequence of Protozoan Forms. Arch. f. Protis. Vol. 65, p. 1.
- ELLIOTT, A. M. 1933. Isolation of *Colpidium striatum* Stokes in Bacteria Free Cultures and the Relation of Growth to pH of the Medium. Biol. Bull. Vol. 65 p. 45.
- FINLEY, H. E. 1930. Toleration of Fresh-water Protozoa to Increased Salinity. Ecology, Vol. 11, p. 337.
- HIRO, F. 1938. On the Resistance of Some Littoral Barnacles to Altered Salinity and Sun Exposure. Botany and Zoology Vol. 6, p. 1686; p. 1851.
- JONES, E. P. 1930. *Paramecium* Infusion histories. I. Hydrogen Changes in Hay and Hay-flour Infusions. Biol. Bull. Vol. 59, p. 275.
- LACKEY, J. B. 1936. Occurrence and Distribution of the Marine Protozoan Species in the Woods Hole Area. Biol. Bull. Vol. 70, p. 264.
- LACKEY, J. B. 1938. A Study of some Ecologic Factors Affecting the Distribution of Protozoa. Ecological Monogr. Vol. 8, p. 501.
- MAST, S. O. 1931. Effect of Salts, Hydrogen-ion Concentration, and Pure Water on Length of Life in *Amoeba proteus*. Physiol. Zool. Vol. 4, p. 58.
- MORGAN, W. DE. 1925. Further Observations on Marine Ciliates Living in the Laboratory Tanks at Plymouth. J. Mar. Biol. Assoc., N. S. Vol. 14, p. 23.
- NOLAND, L. E. 1925. Factors Influencing the Distribution of Fresh Water Ciliates. Ecology, Vol. 6, p. 457.
- OKADA, Y., UYEMURA, M., and ITO, Y. 1939. On the Biological Studies of Thermal Water in Japan. The Zoological Magazine (Tokyo), Vol. 51, p. 39.
- PEARSE, A. S. 1928. On the Ability of Certain Marine Invertebrates to Live in Diluted Sea Water. Biol. Bull. Vol. 54, p. 405.
- PORT, J. 1927. Die Wirkung der Neutralsalz auf die Koagulation des Protoplasmas bei *Paramecium caudatum*. Protoplasma, Bd. 2, S. 101.
- POYARKOFF, E. 1928. La méthode statistique dans la protophysiologie. Protoplasma, Bd. 3, S. 550.
- PRUTHI, H. S. 1927. On the Hydrogen-ion Concentration of Hay Infusions, with Special Reference to its Influence on the Protozoan Sequence. Brit. Jour. Exp. Biol. Vol. 4, p. 292.
- UYEMURA, M. 1939. On the Upper Limit of the Biokinetic Temperature of the Thermophilous Animals found in Hot Springs. Botany and Zool. Vol. 7, p. 1384.
- WORLEY, L. G. 1929. The Marine Rotifer *Brachionus mülleri* subjected to Salinity Changes. Ecology, Vol. 10, p. 420.

SYMBOLAE ITEOLOGICAE VIII¹

AUCTORE

ARIKA KIMURA

Instituto Biologicae Tōhoku Universitatis Imperialis Sendaiensis

Cum 7 tabulis et 7 figuris in textu

Opus acceptum die 9 Sept. 1910

90) ***Salix Kinuyanagi***² KIMURA sp. nov. (Fig. 1, 2 & Tab. X, XI).

Syn. "*Salix viminalis* L." SEEMEN, *Salic. Jap.* p. 50 (1903) pro parte, quoad specim. ex Tokyo (SHIRAI n. 15). MATSUMURA, *Ind. Pl. Jap.* II: 2, p. 15 (1912) pro parte, quoad specim. ex Tsu & Tokio. MAKINO & NEMOTO, *Cat. Jap. Pl. Herb. Nat. Hist. Dept. Tokyo Imp. Mus.* p. 311 (1911). ONUMA n SHIRAI & ONUMA, *Honzozuhu Meiso ad TUNEMASA IWASAKI, Honzozuhu LXXXI.* ed. ann. 1920.

Salix viminalis var. *yezoensis* SCHNEIDER in SARGENT, *Pl. Wilson.* III, p. 158 (1916) pro parte, quoad pl. ex prov. Musashi (H. WILSON n. 6119).

Salix Schuerinii WOLF var. *Kinuyanagi* KIMURA in sched.

Descr. lata: *Frutex* vel *arbor* usque 5-6 m. alta, coma rotundata, trunco cortice sordide cinereo vetustate longitudine irregulariter rimoso obtecto. *Ramuli* annotini longissimi rectiusculi aliquantum crassi, hieme olivaceo-virides vel castaneo-virides, pilis cinereis velutino-tomentosi, inferne vestigiis tomenti residuis tecti aut glabrescentes, tunc plus minus nitiduli; rami cicatricosi cortice griseo-olivaceo, interiore luteo-viride. *Gemmae* amentiferae adpressae, in ramulis eximie condensatae, ovato-vel oblongo-ellipticae vel anguste ovatae, apice obtusissimae latere obtuse carinatae, sordide stramineae vel cinereo-helvolae, pilis cinereis velutino-sericeae, (7-)10.5-12(-13.5) mm longae (3.0-)1.0(1.5) mm latae, 2.6-3.0-plo longiores quam latiores; foliiferae adpressae, stramineae vel brunneae, aliae in supremis partibus ramuli locatae, oblongae vel elliptico-oblongae, apice obtusissimae, pubescentes, 5-7 mm longae et circiter 2.5 mm latae, aliae inferne vel rarius inter amentiferas sitae et ellipticae vel ovatae, apice

¹ Opusculum hoc partim effectum per subsidium mihi donatum ab Nippon-Gakuzvutu-Sinkokai sive Societate Japonica pro Promotu Investigationum Scientiarum.

² Ex nomine vernaculo; kinu-sericea, yanagi-salix, propter folia subtilis sericea. Specimina hujus speciei antea emisi sub nomine "*Salix serotina* PALLAS var. *Kinuyanagi* KIMURA" ad herbarium Arboreti Arnoldiani et R. Museum Historiae Naturalis Holmiae.

obtusissimae, velutinae, 5-7 mm longae, 2.5-3.0 mm latae, demum interdum glabrescentes. *Cataphylla* sterilium ramulorum lineari-oblonga utrinque



Fig. 1. *Salix Kinuyanagi* KIMURA. A Ramuli pars amentiferi sub anthesi. $\times 1$
B Ramuli pars cum gemmis amentiferis $\times 1$.

obtusa, margine integerrima, supra inferne pilosiuscula ceterum glabra, subtus adpresse argenteo-villosissima, 11-20 \times 2.5-4.0 mm magna. *Folia recentissima* utrinque (subtus satis densius) sericea, e vernatione relaxata margine (supremo excepto) evidenter revoluta; *adultae* chartacea internodiis 3-11 mm longis dissita, anguste lanceolata, apicem versus attenuato-acuminata, basi margine leviter convexo acuta vel obtusa, 10-19.5 cm longa 1.1-1.9 cm lata, circiter 9-12-plo longiora quam latiora, margine leviter revoluta, obsolete glanduloso-crenato-serrulata, serraturis 0.8-2.8 mm inter se distantibus, supra saturate viridia sub lente inaequaliter sparse adpressequae puberula, demum glaberrima paullo nitida, subtus pilis rectis adpressis densissimis 0.5-1.0 mm longis costaque parallelis argenteo micantia;

costa straminea vel rubicunda supra elevata minute pubescente, subtus valde prominente holosericea; nervis primariis supra leviter impressis subtus prominulis, utrinque 23-30 a costa sub angulis 40° - 80° exeuntibus, arcuatis ante marginem adscendentibus leviterque flexuosis, secundariis irregularibus infra pube densissima fere invisibilibus, intermediis 1-3. *Petoli* supra superne convexi basin versus sulcati, ad basin dilatati, undique sericeo-tomentosi, 1.0-1.8 cm longi. *Stipulae* oblique lanceolatae vel oblique ovato-lanceolatae, apice attenuato-acuminatae, basi semicordatae, margine plus minus revolutae, obsoletissime crenato-serrulatae, supra convexae sericeae, subtus concavae pilis densis adpressis argenteo-micantes, costa media prominente, nervis prominulis, 11-20 mm longae 3-5 mm latae. *Amenta* ♂ praecocia in ramulis eximie condensata, ellipsoidea vel ovoidea, recta brevia densiflora, apice obtusissima vel rotundata, basi sessilia cataphyllis suffulta, rhachidibus sericeis, 2.7-3.5 cm longa, 1.6-2.0 cm crassa. *Cataphylla amentorum* 2-6, lanceolata vel lineari-lanceolata, minora satis squamosa et basi excepta nigrescentia, utrinque (infra densius) sericeo-villosa, $4.5-8 \times 1.3-1.8$ mm magna, majora viridia, supra glabra vel partim sericea, subtus sericeo-villosa, $10-14 \times 2-3$ mm magna. *Bracteolae* lanceolato-oblongae, apice acutae, dimidia superiore nigrae, medio albiae vel rubicundae, basi pallide flavo-virides, extus villosae, intus dimidia superiore villosae et basi glabrescentes, 3-3.2 mm longae, 1.0-1.1 mm latae. *Glandula* una ventralis linearis, apice truncata, leviter curvula lutea 1.2-1.6 mm longa circiter 0.4 mm lata. *Stamina* 2, filamentis

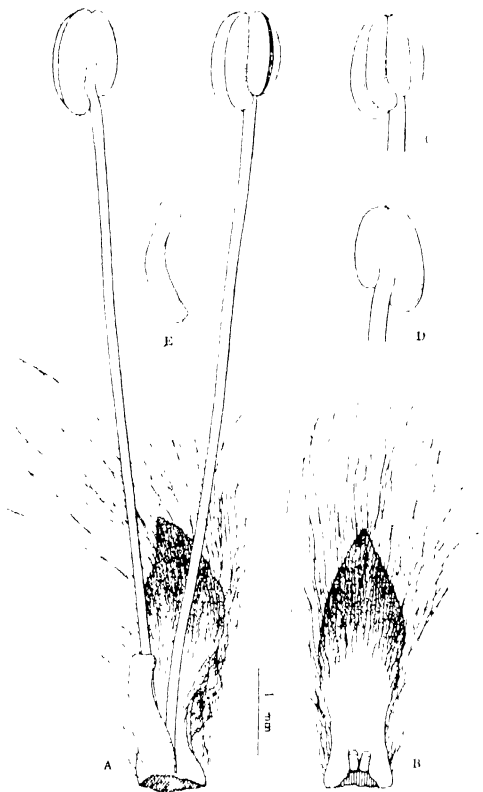


Fig. 2. *Salix Kinuyanagi* KIMURA. A Flos ♂. B Bracteola a facie. C Anthera a facie. D Eadem a dorso. E Glandula ventralis a latere.

albis glabris liberis filiformibus 7.5–9.0 mm longis. Antherae luteae oblique ovoides, $1.2-1.3 \times 0.8$ mm magnae. Planta ♀ ignota.

Nom. Jap. *Kinuyanagi* TUNEMASA IWASAKI¹, Honzozuhu LXXXI. fol. 17 verso (1828) mss.

Hab. in Japonia (cult.). Honsyû. — Prov. **Rikuzen**: Sendai, (A. KIMURA n. 2192 ♂ fl. [typus] 11 Apr. 1933 in Herb. A. KIMURA, fol. [typus fol.] 18 Oct. 1931). — Prov. **Hitati**: Midorigaokamura, (TURUMATI n. 5 fl. 25 Mart. 1930, fol. 15 Jun. 1930). — Prov. **Musasi**: in planitie Simura, (A. KIMURA n. 2852 fol. 25 Maio 1929); prope oppidum Siki, (A. KIMURA n. 2853 fol. 30 Oct. 1923); Tokyo-Sugamo, (A. KIMURA n. 2854 fol. 2 Nov. 1923); Tokyo-Nakano, (A. KIMURA n. 21 fol. 6 Nov. 1923). — Prov. **Etizen**: Turuga, (Y. HOSOI n. 26 fol. 19 Aug. 1936). — Prov. **Settu**: in ripis fl. Kanzakigawa, (A. KIMURA n. 2855 fol. 8 Sept. 1923). — Prov. **Inaba**: Tunoimura, (Y. IKOMA n. 3 gemm. 5 Jan. 1933). — Prov. **Bingo**: inter Tôzyô et Sinryûko, (K. NAOHARA fol. 1 Sept. 1931). — Prov. **Suo**: Kagawa, (T. ODA n. 3172 fol. 9 Sept. 1932); Hirakawa, (T. ODA n. 3179 fol. 9 Sept. 1932); Yamaguti, (T. ODA gemm. 10 Dec. 1932, Feb. 1933).

Sikoku. — Prov. **Tosa**: Higasiyamamura-Yasunami, (T. MORIMOTO n. 9 fol. 17 Aug. 1935).

Kyûsyû. — Prov. **Hyûga**: Miyazaki, (T. HINO n. 52 fl. 12 Mart. 1933). — Prov. **Buzen**: “Kiushu, in humidis littoris Bakan,” (U. FAURIE n. 5350 fol. Jul. 1903 in Herb. Univ. Imp. Kyotensis).

A *Salice Schwerinii* WOLF atque *S. Pet-susu* KIMURA, quibus maxime affinis, potissime differt ramulis crassioribus tomento densiore tectis, gemmis amentiferis majoribus confertius locatis, foliis subtus pube densiore validius argenteo-micantibus. — Nobis haec salix nonnisi e stirpe masculina cognita est. Colitur ubique in insulis Kyûsyû, Sikoku, Honsyû media et australi (boreali autem rarissime) ad pagos et circa domos, sed nequaquam sponte invenitur; fieri potest, ut ex Korea antiquis temporibus primo in Kyûsyû vel occidentalem partem Honsyûensem introducta et deinde boreali-orientalem usque distributa sit. TUNEMASA IWASAKI (1786–1842), ille illustrissimus botanicus Yedoensis, in suo magno opere *Honzozuhu* mentionem fecit salicem hanc in provincia Mino occurrere. Ista mentio suggerit mihi ut haec suo tempore nondum in Yedo culta esset, nunc autem ibi et vicinitate haud rara est.

¹ Cf. E. D. MERRILL & E. H. WALKER, A Bibliography of Eastern Asiatic Botany p. 214 (1938). •

² Per benevolentiam dom. SINZIRO KYÔDÔ Sendaiensis specimen originale amplexu cotypica ex stirpe in horto suo culta legi; illi gratias ago.

91) *Salix Kingoi*¹⁾ KIMURA sp. nov. (Fig. 3 & Tab. XII).

Descr. lata: — *Frutex* humilis prostratus circiter 15–25 cm altus. *Ramuli* annotini 7–18 cm longi, torulosi glaberrimi nitiduli, sub prelo plerumque fuscescentes vel atro-fuscescentes et interdum dilute pruinati, internodiis in medio ramuli 1.5–3.5 cm longis; hornotini ab initio glaberrimi. *Cataphylla* sterilium ramulorum late elliptica ad obovata, apice obtusissima vel rotundata, basi obtusissima vel subcuneata, margine minutissime glanduloso-serrulata, supra puberula vel glabra, subtus secus costam vel antice tantum adpresse villosa vel glabra, prima $4-7 \times 2.5-4.5$ mm, secunda $7-8 \times 4-6$ mm, tertia $9-15 \times 6-9$ mm magna. *Folia juvenilia* supra adpresse sericea infra glaberrima, vernatione convoluta; *adultia* rigide chartacea pleraque obovata interdum elliptica vel ovalia, apice rotundato-acuta vel subcuspidato-rotundata vel fere rotundata nonnumquam acuta vel breviter acuminato-acuta, basi acuta vel subcuneatim acuta vel obtusa, margine circumcirca dense minute arguteque glanduloso-serrulata, serraturis in medio folii 6–10 pro 1 cm, supra leviter nitentia viridia non stomatifera, subtus intense glauca, utrinque glaberrima, $3.0-5.6 \times 2.3-3.0$ cm magna,

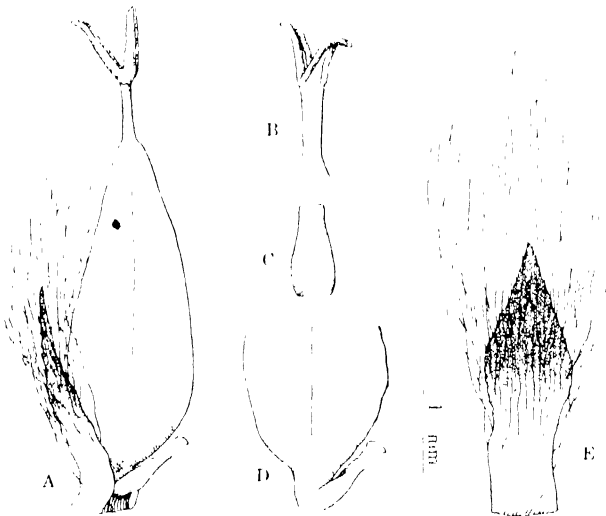


Fig. 3. *Salix Kingoi* KIMURA. A Flos ♀ a latere. B Stigmata cum stylo. C Glandula ventralis a facie. D Basis ovarii cum glandula a latere. E Bracteola a facie.

¹⁾Species dicavi cl. dom. DR. KINGO MIYABE, Academiae Imperialis Scientiarum socio ac Botanici Professori Honorario Imperialis Universitatis Hokkaidensis, qui mihi hanc novitatem ut investigarem misit.

1.5–2.0-plo longiora quam latiora; costa supra fere plana in juventute puberula mox glabrescente, infra prominente glaberrima; nervis primariis leviter arcuatis supra in sicco elevatis infra prominentibus, utrinsecus 9–14, a costa sub angulis 40°–70° divergentibus, superioribus plus minusve acrodromis, secundariis subirregularibus pulchre anastomosantibus. *Petiolis* semiteretes supra sulcati pilosi infra convexi glabri 5–9 mm longi. *Stipulae* forma variae oblique ovatae vel oblique lanceolato-ovatae vel oblique lanceolatae, plerumque superiores majores ac latiores, apice acutae margine serrulatae utrinque glabrae subtus glaucae, 3.4×1.5 , 5×2.5 , 7×3.5 , 8×5 , 10×4 , 9×2.5 mm etc. magnae. *Amenta* ♀ (semimatura et fructifera visa) coactanea recta erecta oblongo-cylindrica densiflora, semimatura 3.5–6.0 cm longa, 1.0–1.3 cm crassa, fructifera ad 9 cm longa, rhachidibus pilosis, pedunculis 6–11 mm longis pubescentibus vel villosis; cataphylla pedunculi 2–3 squamosa, florum bracteolis fere conformia et fere aequimagina, dimidio superiore fusco-atra, utrinque dense albo-villosa, rarius quorum unum majus ellipticum foliaceum viride. *Bracteolae* oblongae apice acutae vel obtusae, dimidia superiore fusco-atrae, utrinque villosae 2–3 mm longae circiter 1 mm latae. *Glandula* una ventralis ovato-oblonga vel oblonga truncata 1 mm longa 0.4–0.5 mm lata. *Ovaria* (semimatura visa) ex ovata basi longe conica acuta glaberrima 3.5 mm longa, stipitibus glabris vel paucipilosis 0.3–0.4 mm longis; stylis obcompressis 0.7–0.8 mm longis. *Stigmata* commissuralia linearia divaricata, apice integra vel leviter emarginata vel bifida 0.5–0.8 mm longa. Capsulae stramineae circa 7 mm longae 2.5–3.0 mm crassae. Stirps ♂ mihi ignota.

Hab. in Japonia. Sachalin austr. — Distr. **Sikka**: monte Kawasimayama, (TATEWAKI & TAKAHASI n. 22754 ♀ [typus] 23 Jun. 1936 in Herb. Hokkaido Univ. Imp.; n. 22751 ♀ 23 Jun. 1936 in Herb. Hokkaido Univ. Imp.; n. 22682 ♀ 21 Jun. 1936 in Herb. Hokkaido Univ. Imp. — S. SUGAWARA n. 28286 ♀, n. 28289 ♀ 3 Aug. 1935 in Herb. A. KIMURA; n. 28285 ♀, n. 28303 ♀, n. 28294 ♀, n. 28299 ♀, n. 28290 ♀, n. 28295 ♀ 3 Aug. 1935 in Herb. Hokkaido Univ. Imp. — M. KAWASHIMA n. 2 ♀ “In alpine meadow: Chirikoro”, 16 Jul. 1935 in Herb. Hokkaido Univ. Imp.).

Rami procurentes, amenta lateraliter erecta pedunculata, foliorum forma et serratura affinitatem cum *S. Chamissonis* demonstrant, a qua tamen bene differt: habitu robustiore, foliis majoribus crassioribus subtus sat glaucis (nec pallidioribus) minus dense serrulatis, stipulis majoribus latioribus, amentis brevius pedunculatis, capsulis stramineis (nec violaceo-fuscis) multo brevius stipitatis (stipitibus in *S. Chamissonis* ad 1 mm longis)

stigmatibus distincte commissuralibus (nec carinalibus). Examinaui bonam materiam *S. Chamissonis* ex sinu S. Laurentii, loco classico, per benevolentiam cl. dom. Dr. K. MIYABE, cui gratias ago.

form. **hebecarpa** KIMURA f. nov.

A typo recedit ovariis superne pubescentibus.

Descr. specim. original.: Flores ♀ paullo post anthesin: *bractcolae* oblongae apice obtusissimae dimidio superiore fusco-atrae utrinque villosae 2 mm longae vix 1 mm latae. *Glandula* una ventralis ovata truncata 0.8 mm longa 0.5 mm lata. *Ovaria* lanceolato-conica 3.5 mm longa dimidia superiore parte pubescentia, ima basi et secus lineam suturalem pilosa; stipitibus 0.5 mm longis pilosis; stylis 0.8 mm longis. *Stigmata* 0.8 mm longa divaricata plerumque bilobata.

Hab. in Japonia. Sachalin austr. — Distr. **Sikka**: Naruko-Nirayama, (TATEWAKI & TAKAHASI n. 22866 ♀ [typus form.] 26 Jun. 1936 in Herb. Hokkaido Univ. Imp.; n. 22866' ♀ 26 Jun. 1936 in Herb. Hokkaido Univ. Imp.); monte Kawasimayama, (TATEWAKI & TAKAHASI n. 22679 ♀ 21 Jun. 1936; 22752 ♀ 23 Jun. 1936; n. 22573 ♀ 23 Jun. 1936 in Herb. Hokkaido Univ. Imp.).

form. **macrocataphylla** KIMURA f. nov.

A typo recedit pedunculis amentorum cataphyllis majoribus foliaceis praeditis.

Descr. specim. original.: — *Amenta* ♀ oblongo-cylindrica densiflora sub anthesi 2-3.2 cm longa circiter 1 cm crassa. Pedunculi 0.9-1.4 cm longi villosi ima tantum basi glabri. *Cataphylla* pedunculorum 3-4, elliptico-vel obovato-elliptica, apice obtusa vel acuta basi cuneato-acuta, breviter petiolata, margine minute serrulata, supra glabra subtus glauca glabra vel inferne ad costam villosa, majora ad 1.8 cm longa 0.7 cm lata; sterilium ramulorum fere ut pedunculorum. Flores ut in typica.

Hab. in Japonia. Sachalin austr. — Distr. **Sikka**: monte Kawasimayama, (TATEWAKI & TAKAHASI n. 22681 ♀ [typus form.] 21 Jun. 1936 in Herb. Hokkaido Univ. Imp.; n. 22686 ♀ 21 Jun. 1936; n. 22685 ♀ 21 Jun. 1936 in Herb. Hokkaido Univ. Imp.).

92) × **Salix Koiei**¹⁾ KIMURA hyb. nov. (Fig. 4 & Tab. XIII, XIV).

= *Salix Gilgiana* SEEMEN × *S. gracilistyla* MIQUEL.

Descr. specim. original.: — *Frutex* circiter 1.5 m altus habitu fere *S.*

¹⁾ In honorem collectoris nominata.

Gilgianae (ex collectore). *Ramuli* hornotini teretes molliter griseo-tomentosi deinde glabrescentes; annotini elongati in sicco luteo-brunnei vel fuscescentes glabri vel superne vestigiis tomenti residuis obducti. *Gemmae* amentiferae (in ramulis auctumno lectis visae) ovato-ellipticae obtusae latere carinatae ventre moderatim dorso valde convexae, primo sericeo-tomentosae demum glabrescentes, in sicco brunneae 1.5 cm longae 0.5 cm latae; foliiferae ovatae sericeae vel glabrescentes ad 6 mm longae. *Folia adulta* chartacea internodiis 1.4–2.6 cm longis dissita, oblonga ad lanceolato-oblonga, medio fere latiora, apice acuminata basi obtusissima vel rotundata,

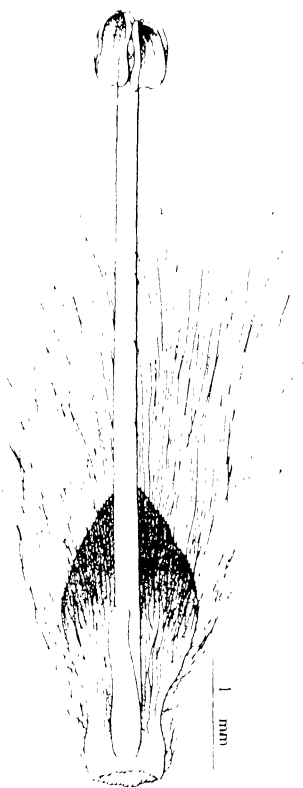


Fig. 1. *Salix Koiei* KIMURA.
Flos masculinus.

marginē anguste reflexa glanduloso-crenato-serrulata, serraturis in medio folii 3–4 pro 1 cm sursum crebris, acumine et basi integra, supra viridia glabra stomatifera subtus glauca pilis minutis adpressis et sparse et dense pubescentia, 10–13.5 cm longa 2.4–3.3 cm lata, 3.5–1.9-plo longiora quam latiora; costa supra elevata pulverulento-pubescente demum glabrescente, subtus valde prominente sericeo-pubescente demum glabrescente; nervis primariis arcuato-ascendentibus utrinsecus 13–15 a costa sub angulis 50°–60° divergentibus supra in sicco fere planis vel elevatiusculis infra prominentibus, secundariis tenuibus subparallelis crebris inter primarios transversis; intermediis 2–3. *Petioles* semiteretes supra basi sulcati, primo circumcirca tomentosi, creta gemma subtus glabrescentes ad 1 cm longi. *Stipulae* oblique ovatae acuminatae glanduloso-serrulatae utrinque puberulae supra ima basi glandulosae infra glaucae ad 12 mm longae 5 mm latae. *Amenta* ♂ praecocia oblongo-cylindrica apice obtusissima curvula densiflora sessilia ante anthesin villosissima, rhachidibus pubescentibus, 3.5–4.8 cm longa ad 1.5 cm crassa, basi plerumque

cataphyllis paucis squamosis anguste deltoideis vel ovato-oblongis acutis vel obtusis integerrimis utrinque villosis vel supra antice glabrescentibus 5.5–7 × 2.2–2.5 mm magnis suffulta. *Bracteolae* ellipticae vel subrhombello-ellipticae vel fere obovatae sursum acutae ad summum obtusae vel acutae

deorsum subcuneatae et intus concavae, utrinque albo-villosissimae, dimidia superiore nigrae inferiore pallidae 2.6-3.0 mm longae circiter 1.3 mm latae. *Glandula* una ventralis linearis truncata paullo incurvata 1.1-1.3 mm longa. *Stamina* 2, filamentis glabris prorsus ad apicem usque connatis 5.3-5.6 mm longis. *Antherae* ovae saltem apice purpureae 0.6-0.9 mm longae effuso polline plerumque fuscescentes.

Hab. in Japonia. Honsyû. — Prov. Ugo: Huzikoto-mura, (G. KOIE n. 34 ♂ fl. [typus] 21 Apr. 1910 in Herb. A. KIMURA, fol. [typus fol.] 2 Oct. 1939; n. 35 ♂ fl. 25 Apr. 1910, fol. 2 Oct. 1939).

Magis tendet ad *S. gracilistylam*: ramulorum tomento, figura et magnitudine gemmarum, forma nervatura serratura indumento foliorum, forma stipularum, figura et magnitudine amentorum, villositate densa bracteolarum, glandula elongata et glabritate filamentorum. Bracteolarum autem forma certissime mixturam cum *S. Gilgiana* demonstrat.

93) × *Salix thaymasta* KIMURA in Tokyo Bot. Mag. LX. p. 611 (1926). — MAKINO & NEMOTO, Fl. Jap. ed. 2, p. 172 (1931). — NEMOTO, Fl. Jap. Suppl. p. 117 (1936). — HONDA, Nom. Pl. Jap. p. 15 (1939).

= *Salix gracilistyla* MIQUEL × *S. Kinuyanagi* KIMURA.

Fig. 5. Descr. ♂: *Frutex* circiter 2 m altus. *Ramuli* teretes elongati, annotini pilis cinereis velutino-tomentosi, inferne saepe glabrescentes paullo nitentes, in sicco sordide lutei vel luteo-brunnei, inferne 4 mm superne 2 mm crassi, vetustiores fuscescentes. *Gemmae* amentiferae non visae; foliiferae stramineo-brunneae, ovato-ellipticae, apice obtusae, cinereo-sericeae vel glabrescentes, 4-6 mm longae, 1.5-2.5 mm latae. *Folia recentissima* utrinque dense sericea, e vernatione relaxata margine evidenter revoluta; *adultae* chartacea interstitiis 5-16 mm longis dissita, lanceolata, apice sensim acuminata basi acuta, 8-10 cm longa, 1.2-1.6 cm lata, 5.6-8-plo longiora quam latiora, margine leviter revoluta glanduloso-crenato-serrulata, serraturis in medio folii 5-7, superne minutis sed acutis 14-16 pro 1 cm, basin versus laxantibus et

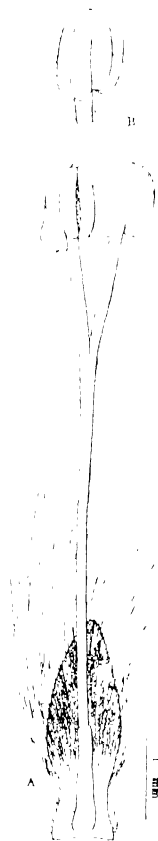


Fig. 5. *Salix thaymasta* KIMURA.
A Flos ♂. B Anthera a facie.

obsoletis, supra viridia inaequaliter adpresseque puberula (demum glaberrima?), subtus pilis adpressis acroscopicisque dense sericea; costa supra elevata minute pubescente, subtus valde prominente dense sericea; nervis primariis supra leviter impressis subtus prominentibus, utroque latere 18–20 a costa sub angulis 30° – 50° arcuato-ascendentibus, secundariis crebris inter primarios transversis, infra pube densa fere invisibilibus, intermediis 1–3. *Petoli* supra sulcati circiter 1.0 cm longi undique sericeo-tomentosi. *Stipulae* oblique ovato-lanceolatae acuminatae minute serrulatae ad 15 mm longae 4 mm latae, supra sericeae, subtus concavae pilis adpressis densius sericeae. *Amenta* ♂ praecocia densiflora oblongo-cylindrica sessilia, rachidibus pubescentibus, 2.5–4 mm longa circiter 1.5 cm crassa, basi cataphyllis squamosis sessilibus 1–3 vel nullis lineari-oblongis apice obtusis supra glabris subtus sericeo-villosis margine integerrimis 4.5–6 mm longis 1.3–1.8 mm latis suffulta. *Bracteolae* lanceolato-oblongae superne acutae ad summum obtusae, circiter 2.5 mm longae 0.8 mm latae, basi pallidae ceterum nigrae, utrinque albo-villosae. *Glandula* una ventralis lutea anguste linearis, apicem versus paullo attenuata, apice rotundato-truncata, 1.3–1.6 mm longa inferne circiter 0.3 mm lata. *Stamina* 2, filamentis albis glabris 6–7 mm longis, e basi ad $2/3$ – $6/7$ totae longitudinis connatis. *Antherae* ovales circiter 1 mm longae, apice rubrae basi luteae, post anthesin fusciscentes.

Hab. in Japonia. Honsyû. — Prov. **Settu**: monte Rokkusan, (E. ISIKAWA ♂ fl. [typus ♂] 8 Apr. 1940; fol. 23 Aug. 1939). Flores et folia ex una eademque stirpe!

Medium inter *S. gracilistylam* et *S. Kinuyanagi* fere tenet. ita ut facile pro hybrida earum haberi posset. Ab illa habet foliorum nervationem (i. e. nervos secundarios crebros inter primarios transversos), serraturam haud obsoletam, glandulam floris anguste linearem, filamenta staminum alte connata, sed a *S. Kinuyanagi* folia adulta lanceolata sursum attenuata subtus dense sericea, recentissima e vernatione relaxata margine evidenter revoluta.

94) × **Salix ampherista** SCHNEIDER in SARGENT, Pl. Wilson. III. p. 175 (1916). — MAKINO & NEMOTO, Fl. Jap. ed. 2, p. 161 (1931). — KIMURA in Jour. Fac. Agr. Hokkaido Imp. Univ. XXVI. 4, p. 452 (1934) (MIYABE & KUDO, Fl. Hokkaido & Saghal. IV.). — NEMOTO, Fl. Jap. Suppl. p. 105 (1936). — HONDA, Nom. Pl. Jap. p. 41 (1939).

= *Salix sachalinensis* SCHMIDT (!) × *S. vulpina* ANDERSSON (?).

Fig. 6 & Tab. XV, XVI. — Descr. specim. original.: — *Ramuli* annotini

tenuis ad 18 cm longi basi 2 mm superne 1 mm crassi glaberrimi purpureo-fusci paullo nitentes, hornotini brunnei minute pubescentes. *Cataphylla* sterilium ramulorum oblonga obtusa integerrima brevissime petiolata supra glabra subtus adpresse villosa 10-11 mm longa 3.5-5.0 mm lata. *Folia* (adulta nondum evoluta) *intermedia* tenuia late elliptica, obovata, obovato-elliptica, apice late acuta ad summum obtusa, basi acuta vel subcuneatim angustata, margine integerrima, 3.5 × 2, 3.6 × 2.2, 1.8 × 2 cm etc. magna, supra viridia sparse adpresse puberula, subtus pallidiora sparse adpresse sericea; costa supra fere plana vel subimpressa minutissime puberula infra prominula adpresse pilosa; nervis primariis infra vix elevatis utrinque 11-11 a costa sub 40°-70° divergentibus, secundariis subregularibus. *Folia recentissima* utrinque sericeo-pubescentia, e vernatione relaxata margine infero revoluta ad modum *S. sachalinensis*. *Petoli* foliorum intermediorum semiteretes supra sulcati circumcirca minutissime pubescentes ad 6 mm longi. *Amenta* tantum fructi-

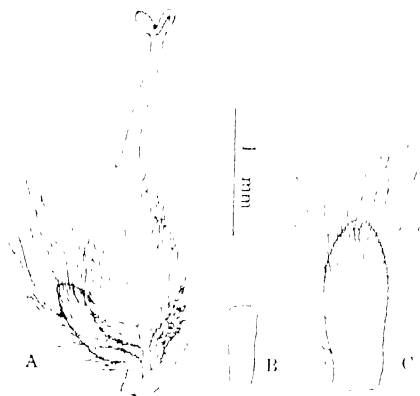


Fig. 6. *Salix ampherista* SCHNEIDER. A Flos ♀ a latere. B Glandula ventralis. C Bracteola a facie, fere expansa.

fera visa, ut videtur praecocia, brevissime pedunculata, pedunculis villosis circiter 4 mm longis, 3-5.5 mm longa circiter 1.2 cm crassa, basi cataphyllis paucis ellipticis obtusis integerrimis supra glabris infra adpresse villosis circiter 10 mm longis 3-5 mm latis suffulta, rhachidibus sericeo-villosis. Flores inter fructus remanentes: *bracteolae* oblongae apice obtusissimae supra basin constrictae (modo *S. sachalinensis*) 1.3-1.4 mm longae 0.5-0.6 mm latae, dimidia superiore brunneae, utrinque (intus superne tantum) albo-villosae, intus concavae (modo *S. sachalinensis*). *Glandula* una ventralis oblonga truncata compressa 0.6-0.7 mm longa, stipitibus duplo longior. *Ovaria* ovato-conica acuta breviter sericea 1.5-1.8 mm longa; stipitibus pilosis vix 0.3 mm (in fructu vix 0.5 mm) longis; stylis obcompressis 0.8 mm longis integris. *Stigmata* breviter ovata commissuralia integra vel leviter emarginata 0.2 mm longa. Capsulae 5 mm longae. Ovula in quaque placenta 3.

Hab. in Japonia. Hokkaido. Prov. **Osima**: Hakodate, (FAURIE n.

5758 fr. [typus] 1 Jun. 1901, sub nomine *S. daiseniensis* SEEMEN in Herb. Arboreti Arnoldiani).

De origine hujus mirae formae nondum aliquid ad persuadendum dicere possum; propius autem accedit ad *S. sachalinensem* cataphyllorum figura vestimento et magnitudine, foliis intermediis integerrimis subtus pallidioribus sparse sericeis, nervis primariis distinctis et crebris, secundariis subregularibus, foliis juvenilibus utrinque pubescentibus e vernatione relaxatis margine infero revolutis, florum bracteolis prope basin constrictis, ovariis ovato-conicis breviterque sericeis, stylis elongatis, stigmatibus parvis et commissuralibus; sed ab ea praecipue recedit foliis intermediis multo latioribus, glandula ventrali brevior, ovariis brevius stipitatis, quae notae forsitan ex *S. vulpina* derivatae essent. Hic gratias maximas agere mihi est officium cl. dom. Prof. A. REHDER Arboreti Arnoldiani, quod mihi materiam originalem libenter commodavit.



Fig. 7 Typus *Salicis kakistae* SCHNEIDERI.
E. H. WILSON n. 7103.

95) *Salix Reinii* FRANCHET & SAVATIER, Enum. Pl. Jap. I. p. 159 (1875) (nomen). SEEMEN, Salic. Jap. p. 41, t. 6, f. A E (1903) (descr.). KOIDZUMI in Tokyo Bot. Mag. XXVII. p. 91 (1913). SCHNEIDER in SARGENT, Pl. Wilson. III. p. 127 (1916). KIMURA in Jour. Fac. Agr. Hokkaido Imp. Univ. XXVI. 1, p. 402 (1931) (MIYABE & KUDO, Fl. Hokkaido & Saghal. IV.) excl. syn. *Salix daiseniensis* SEEMEN.

Syn. "*Salix glabra* SCOPOLI" FRANCHET & SAVATIER, Enum. Pl. Jap. II. 1, p. 503 (1876).

"*Salix Sieboldiana* BLUME" MATSUMURA, Nippon Shokubutsumei

p. 170 (1884); Cat. Pl. Herb. Imp. Univ. p. 181 (1886) pro parte, excl. pl. ex Siomitôge prov. Kii; Shokubutsu Mei-I p. 261 (1895).—MIYOSHI in Tokyo Bot. Mag. V. p. 88 (1891).—MATSUDAIRA & IKENO in Tokyo Bot. Mag. VIII. p. 425 (1894).—ICHIMURA in Tokyo Bot. Mag. XIII. p. 101 (1899). **Syn. nov.**

“*Salix Miquelii* ANDERSSON” OKUBO, Cat. Pl. Bot. Gard. Imp. Univ. p. 201 (1887).—MATSUMURA in Tokyo Bot. Mag. III. p. 249 (1889); List Pl. Nikko p. 28 (1894).—WATANABE & MATSUDA in Tokyo Bot. Mag. VI. p. 91 (1892). **Syn. nov.**

Salix kakista SCHNEIDER in SARGENT, Pl. Wilson. III. p. 128 (1916) quoad holotypum. **Syn. nov.** (Fig. 7 nostra).

Nom. Jap. *Iwayanagi* MATSUMURA, Nippon Shokubutsumei p. 170 (1884).—MIYOSHI in Tokyo Bot. Mag. II. p. 199 (1888).

Miyamaynagi MATSUMURA in Tokyo Bot. Mag. III. p. 249 (1889).

Mineyanagi TOKUBUCHI in Tokyo Bot. Mag. X. p. 123 (Apr. 1896).—KAWAKAMI in Tokyo Bot. Mag. X. p. 50 (Jun. 1896).

Salicis kakistae holotypus, quem ipse examinavi, ab pura *S. Reinii* haud differre mihi videtur; itaque illud nomen ut synonymum in hoc reduxi.

var. *eriocarpa* KIMURA in Sci. Rep. Tôhoku Imp. Univ. 4 ser. Biol. VI. p. 190 (1931).—NEMOTO, Fl. Jap. Suppl. p. 114 (1936).—HONDA, Nom. Pl. Jap. p. 44 (1939).

Syn. *Salix kakista* SCHNEIDER in SARGENT, Pl. Wilson. III. p. 128 (1916) quoad specim. FAURIE n. 5752 “Nippon, in sylvis Akita”, 18 Jun. 1905. **Syn. nov.**

Examinavi materiam FAURIEanam in Herbario Arboreti Arnoldiani conservatam.

96) *Salix vulpina* ANDERSSON in Mem. Am. Acad. Arts Sci. n. ser. VI. p. 452 (1859) (GRAY, Bot. Jap.).—SEEMEN, Salic. Jap. p. 37, t. 5, f. F-I, (1903) excl. syn. pro parte.—MATSUMURA, Cat. Pl. Herb. Coll. Sci. Imp. Univ. p. 182 (1886); Ind. Pl. Jap. II: 2, p. 15 (1912) pro parte.—KOIDZUMI in Tokyo Bot. Mag. XXVII, p. 89, 264 (1913).—SCHNEIDER in SARGENT, Pl. Wilson. III. p. 130 (1916).—KIMURA in Jour. Fac. Agr. Hokkaido Imp. Univ. XXVI. 4, p. 404 (1934) (MIYABE & KUDO, Fl. Hokkaido & Saghal. IV.).

Syn. “*Salix Sieboldiana* BLUME” MIYOSHI in Tokyo Bot. Mag. V. p. 158 (1891). **Syn. nov.**

Salix daiseniensis SEEMEN, Salic. Jap. p. 66 (1903) pro parte, quoad

pl. ex Rebunsiri, FAURIE n. 3712. — MATSUMURA, Ind. Pl. Jap. II: 2, p. 9 (1912) pro parte, quoad pl. ex Rebunsiri. **Syn. nov.**

Quum Salicacearum feci revisionem in MIYABE & KUDO, Flora of Hokkaido & Saghalien IV, specimen FAURIEANUM n. 3712 negligenter ad *S. Reinii* FR. & SAV. retuli, quod tamen certe ad hanc ducendum. Examinavi materiam FAURIEANAM in Herbario Universitatis Imperialis Kyotensis. Confer KIMURA in MIYABE & KUDO l. c. p. 403.

97) **Salix pulchroides** KIMURA in Jour. Fac. Agr. Hokkaido Imp. Univ. XXVI. 4, p. 446 (1936) (MIYABE & KUDO, Fl. Hokkaido & Saghal. IV.). — NEMOTO, Fl. Jap. Suppl. p. 113 (1936). — HONDA, Nom. Pl. Jap. p. 44 (1939).

Syn. "*Salix anglorum* CHAMISSE" KUDO in Jour. Coll. Agr. Hokkaido Imp. Univ. XI: 2, p. 96 (1922) (Fl. Paramushir).

"*Salix Chamissonis* ANDERSSON" KOIDZUMI in Tokyo Bot. Mag. XLIV. p. 109 (1930). — MAKINO & NEMOTO, Fl. Jap. ed. 2, p. 162 (1931).

Descr. ♀: — *Frutex* habitu ut in ♂. *Folia adulta* obovata, apice fere rotundata, basi subcuneatim angustata, terminalia subrhombeo-elliptica utrinque acuta, margine apice fere integro excepto crenato-serrulata, serraturis ad medium folii 5–6 pro 1 cm, utrinque glaberrima subtus dilute glaucina, 3.5–3.7 × 1.6–2.3 cm magna; costa supra fere plana infra leviter elevata; nervis primariis leviter arcuatis utrinsecus 6–7, a costa sub angulis 40°–50° divergentibus; secundariis irregularibus. *Petoli* glabri supra sulcati ad 8 mm longi. *Stipulae* lanceolatae obsolete serrulatae ad 4.5 × 1 mm magnae. *Amenta* ♀ (tantum deflorata visa) cylindrica 7.5 cm longa 1.7 cm crassa, rhachidibus pubescentibus, foliato-pedunculata ut in ♂, pedunculis ad 4.5 cm longis, foliis ut in ♂. *Flores* ♀ inter fructus remanentes: *bracteolae* elliptico-oblongae apice obtusissimae utrinque albo-villosae, ad 3 mm longae 1.2 mm latae. *Glandula* una ventralis oblonga vel ovata apice truncata 0.9–1.0 mm longa 0.4–0.5 mm lata. *Ovaria* lanceolato-conica 3.5–4 mm longa supra medium tomentosa; stipitibus 1–1.3 mm (in fructu ad 1.6 mm) longis tomentosis; stylis obcompressis 0.9–1.0 mm longis. *Stigmata* oblonga emarginata (an semper?) 0.8 mm longa.

Hab. in Japonia. Kuriles. — Ins. **Syumusyu**: sine loco speciali indicato, (Y. OKADA [typus ♀] 18 Aug. 1931 in Herb. Hokkaido Univ. Imp.).

98) **Salix melanostachys** MAKINO in herb. Sc. Coll. Imp. Univ. Tokyo ex MAKINO in Tokyo Bot. Mag. XVIII. p. 141 (1904) pro syn. — GÖRZ, Schedae ad fasc. I. Salicac. Asiatic. p. 21 (1931); in FEDDE, Rep. Sp.

Nov. Reg. Veg. XXXII. p. 121 (1933); *ibid.* p. 388 (1933).

Syn. "*Salix subopposita* MIQ." MATSUMURA, Nippon Shokubutsumei p. 170 (1884); Cat. Pl. Herb. Coll. Sci. Imp. Univ. p. 181 (1886). --- OKUBO, Cat. Pl. Bot. Gard. Imp. Univ. p. 202 (1887). **Syn. nov.**

"*Salix purpurea* LINNAEUS" MATSUMURA in Tokyo Bot. Mag. VIII. p. 151 (1894). **Syn. nov.**

Salix gracilistyla MATSUMURA. Shokubutsu Mei-I, p. 260 (1895) pro parte.

Salix Thunbergiana BLUME subsp. *melanostachys* MAKINO in Tokyo Bot. Mag. XVIII. p. 141 (1904).

Salix nigrolepis SHIRAI MS ex MAKINO in Tokyo Bot. Mag. XVIII. p. 141 (1904) pro syn.

Salix gracilistyla MIQUEL subsp. *melanostachys* MAKINO in Tokyo Bot. Mag. XXVIII. p. 175 (1914). -- MAKINO & NEMOTO, Cat. Jap. Pl. Herb. Nat. Hist. Dept. Tokyo Imp. Mus. p. 309 (1914); Fl. Jap. ed. 1, p. 1124 (1925); *ibid.* ed. 2, p. 164 (1931). NEMOTO, Fl. Jap. Suppl. p. 107 (1936).

Salix Thunbergiana BL. var. *melanostachys* MAKINO ex MATSUMURA, Ind. Pl. Jap. II: 2, p. 14 (1912).

Salix gracilistyla, var. *melanostachys* SCHNEIDER in SARGENT, Pl. Wilson. III. p. 164 (8 Maio 1916).

Salix gracilistyla MIQ. var. *melanostachys* MAKINO ex MATSUMURA, Shokubutsu-Mei-I, ed. 9, p. 353 (8 Junio 1916).

Hab. in Japonia. --- Colitur in hortis tantum. ♀ ignota.

99) ***Salix integra*** THUNBERG ex MURRAY, Syst. Veget. ed. 14, p. 880 (1784). -- THUNBERG, Fl. Jap. p. 24 (1784). -- CH. TANAKA in Bull. Sci. Fakult. Terkult. Kjusû Imp. Univ. 1: 4, p. 193 (Sept. 1925). KIMURA in Jour. Fac. Agr. Hokkaido Imp. Univ. XXVI. 4, p. 434 (1931) (MIYABE & KUDO, Fl. Hokkaido & Saghal. IV.). -- TATEWAKI in Trans. Sapporo Nat. Hist. Soc. XVI. p. 78 (1940).

Hab. in Japonia. Kuriles. -- Ins. Etorohu: Naiho, (B. YOSIMURA st. 12 Aug. 1939).

Nova civis florum Kurilensis!

100) ***Salix japonica*** THUNBERG ex MURRAY, Syst. Veget. ed. 14, p. 879 (1784). -- THUNBERG, Fl. Jap. p. 24 (1784); Icon. Pl. Jap. Dec. IV. t. 1 (1802).

var. ***angustifolia*** KIMURA in Tokyo Bot. Mag. XLII. p. 573 (1928). --

MAKINO & NEMOTO, Fl. Jap. ed. 2, p. 166 (1931). NEMOTO, Fl. Jap. Suppl. p. 108 (1936). — HONDA, Nom. Pl. Jap. p. 42 (1939).

form. **Fauriei** (SEEMEN) KIMURA comb. nov.

Syn. *Salix Fauriei* SEEMEN in ENGLER's Bot. Jahrb. XXX. Beibl. LXVII. p. 10 (1901); Salic. Jap. p. 18, t. 8, fig. C-E (1903). — ? LÉVEILLÉ in Bull. Acad. Intern. Géogr. Bot. XIV. p. 209 (1901); XVI. p. 149, 152 (1906). — MATSUMURA, Ind. Pl. Jap. II: 2, p. 9 (1912). — SCHNEIDER in SARGENT, Pl. Wilson. III. p. 134 (1916). KOIDZUMI in Tokyo Bot. Mag. XXX. p. 81 (1916)¹⁾. — MAKINO & NEMOTO, Fl. Jap. ed. 1, p. 1122 (1925); ed. 2, p. 163 (1931). — NEMOTO, Fl. Jap. Suppl. p. 106 (1936). — HONDA, Nom. Pl. Jap. p. 11 (1939). Syn. nov.

A var. *angustifolia* mihi tantum foliis diuturne sericeis recedit.

Hab. in Japonia. Honsyû. — Prov. **Sagami**: "Yamakita, rochers", (U. FAURIE n. 3700 ♀ 8 Maio 1899). — Vidi isotypum in herbario Universitatis Imperialis Kyotensis conservatum.

Salix japonica THUNBERGII solum in Honsyû media (paucis quidem provinciis) invenitur; nominis autem auctor (in Flora sua Japonica pag. 24) eam "iuxta Nagasaki" crescentem dicit. Haec mentio tamen per errorem facta sit.

101) ***Salix neo-fuscata* KIMURA nom. nov.**

Syn. *Salix fuscata* GÖRZ (non PURSH) in GROSSHEIM, Fl. Kavkaza II. p. 7 (1930); in FEDDE, Rep. Sp. Nov. Reg. Veg. XXVIII. p. 122 (1930) (K. KRAUSE, Beiträge zur Flora Kleinasien V: 2); ibid. XXXVI. p. 234 (1934) (Die Gattung *Salix* in Kaukasien). — NASAROV in KOMAROV, Fl. URSS. V. p. 90, 100 (1936).

Hab. in Caucasia: Kanly-su, (MASSALSKY steril.).

102) ***Balsamiflua Denhardtiorum* (ENGLER) KIMURA comb. nov.**

Syn. *Celtis ilicifolia* ENGLER, Pflanzenwelt Ost-Afrikas p. 160 (1895).

Populus euphratica OLIVIER subsp. *Denhardtiorum* ENGLER in Notizbl. Kön. Bot. Gart. Mus. Berlin II. p. 218 (1898); in Sitzungsber. Kön. Preuss. Akad. Wiss. Jahrgang 1904 p. 369 (1904) (Veget. Somaliland); in ENGLER, Bot. Jahrb. XXXVI. p. 252 (1905). — ASCHERSON in Ber. Deutsch. Bot. Gesellsch. XXVI a, p. 358, 360 (1908).

Populus euphratica OLIVIER var. *Denhardtiorum* GOMBOCZ in Math. Termesz. Közl. XXX. p. 72 (1908) (Monogr. Gen. Populi).

¹⁾ Specimina nulla vidi.

Populus Denhardtiorum DODE in Bull. Soc. Dendr. France, 1909, p. 152 (1909); in HOOKER, Ic. Pl. XXXI. t. 3050 (1916). SKAN in D. PRAIN, Fl. Trop. Africa VI:2, p. 325 (1917).

Turanga ilicifolia (ENGLER) KIMURA in Sci. Rep. Tōhoku Imp. Univ. 1 ser. Biol. XIII. p. 387 (1938).

Balsamiflua ilicifolia (ENGLER) KIMURA in Sci. Rep. Tōhoku Imp. Univ. 1 ser. Biol. XIV. p. 192 (1939).

Hab. in Africa tropica.

103) *Salix pentandra* L. sub-p. *pseudopentandra* FLODERUS in Arkiv f. Bot. 20 A, no. 6, p. 57 (1926) [ut (*Salix pentandra* L.) **Salix pseudopentandra* n. subsp.]; ibid. 25 A, no. 10, p. 12 (1933). HULTEN in Kungl. Sv. Vet. Akad. Handl. 3 ser. V:2, p. 17 (1928) (Fl. Kamtch. & Adj. Isl. II.). NASAROV in KOMAROV, Fl. URSS V. p. 206 (1936) in nota.

Syn. *Salix pentandra* LANGSDORFF, Bemerk. auf einer Reise um die Welt etc. p. 231 (1812). CHAMISSE in Linnaea VI. p. 538 (1831) quoad pl. Kamtsch. ERMAN, Verzeich. Thieren & Pflanzen etc. p. 56 (1835) quoad pl. Kamtsch. LÉDEBOUR, Fl. Ross. III. p. 597 (1850) quoad pl. Kamtsch. ANDERSSON in Kongl. Sv. Vet. Akad. Handl. VI:1, p. 35 (1867) (Monogr. Salic.) quoad pl. Kamtsch.; in DE CANDOLLE, Prodr. XVI:2, p. 206 (1868) quoad pl. Kamtsch. HERDER in Act. Hort. Petrop. XI:2, p. 397 (1891) (Pl. Radd. Apet. IV.). WOLF in Act. Hort. Petrop. XXI:2, p. 179 (1903). KOMAROV, Voy. Kamtch. p. 231 (1912); Fl. Penins. Kamtsch. II. p. 7 (1929) quoad syn. NASAROV in KOMAROV, Fl. URSS V. p. 205 (1936) quoad pl. Kamtsch.

Hab. in Japonia, Kuriles. Ins. Etorohu: Syana, (B. YOSIMURA & H. YOKOYAMA fr. 11 Aug. 1938. TAKEDA fl. ♂ & ♀ 18 Jun. 1939. B. YOSIMURA fr. & ♂ defl. 30 Jul. 1939).

Nova civis florae Japoniae!

EXPLICATIO TABULARUM

TAB. X.

Salix Kinuyanagi KIMURA. Typus.
Ramuli amentiferi.

TAB. XI.

Salix Kinuyanagi KIMURA.
Ramulus cum foliis adultis.

TAB. XII.

Salix Kingoi KIMURA. Typus.
Ramuli cum foliis et amentis fructiferis.

TAB. XIII.

Salix Korei KIMURA. Typus.
Ramuli amentiferi.

TAB. XIV.

Salix Korei KIMURA.
Ramuli cum foliis adultis.

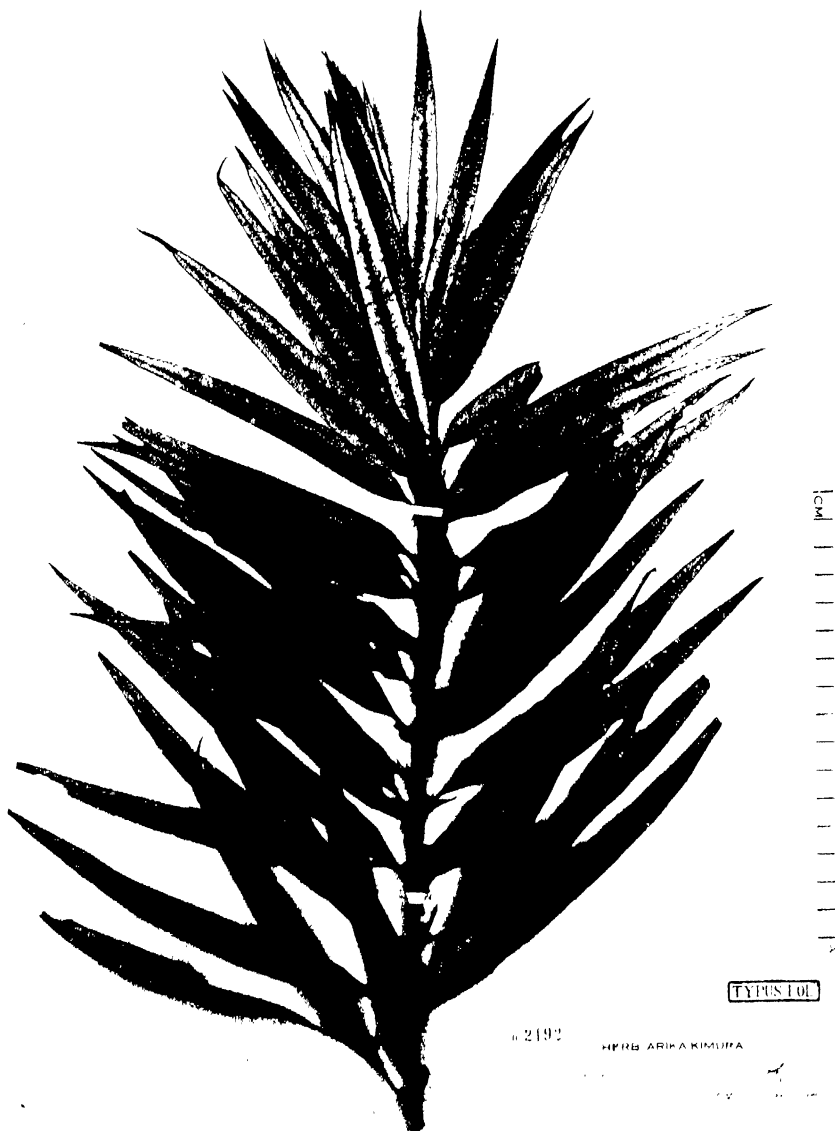
TAB. XV.

Salix ampherista SCHNEIDER. Typus.
Ramuli cum amentis fructiferis et
foliis intermediis.

TAB. XVI.

Salix ampherista SCHNEIDER. Typus. $\times 1$.





n. 2492

HERB. ARIKA KIMURA

In 1930, Kichijiro, Kichijiro, Kichijiro

Leg. Anka Kimura 1.1. Oct. 1939











EMBRYOGENY OF *TORREYA NUCIFERA* S. ET Z.

By

MASATO TAHARA

Biological Institute, Tôhoku Imperial University, Sendai

With Plates XVII, XVIII and 7 text-figures

(Received September 13, 1940)

Torreya, a genus of the family Taxaceae is found only in California, Florida, China and Japan. Embryologically two species have already been studied, namely *T. californica* by Miss ROBERTSON (1904) and *T. taxifolia* by COULTER and LAND (1905). *T. nucifera* is a common tree of Japan, but till now almost nothing has been known regarding its gametophyte and embryo. This year however the material of this plant was collected by the present writer, and the main result of investigation is now presented.

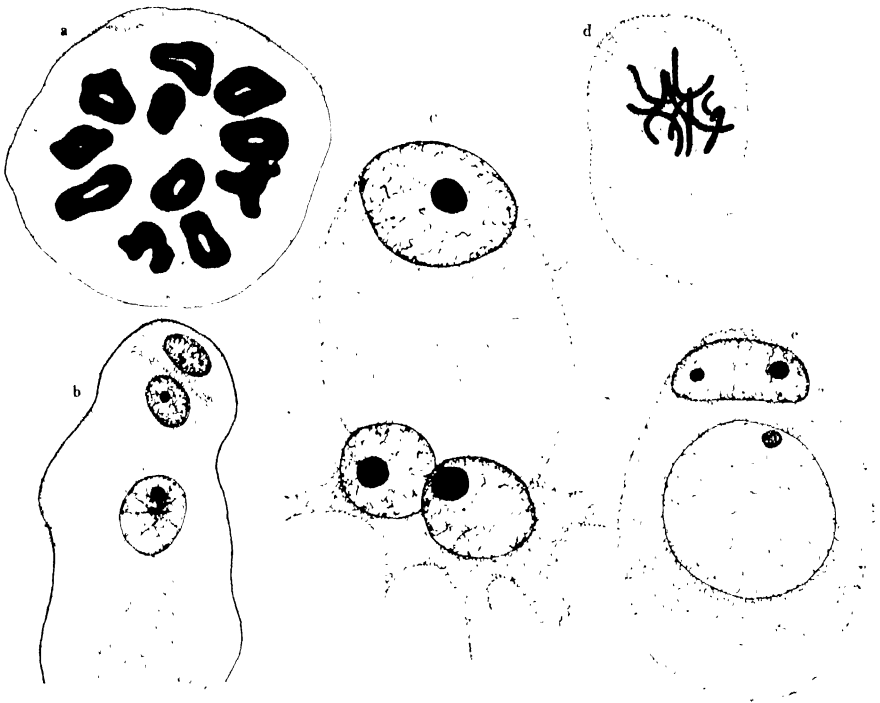
The material was fixed by NAVASHIN's solution after dipping it into formalin-acetic-alcohol (formalin 5, glacial acetic 5, 70% alcohol 90) for ten minutes. Fixation by chromacetic solution did not give a good result in this plant. Sections mostly 15 μ in thickness were stained with HEIDENHAIN's iron-alum haematoxylin. For the study of embryogeny clear staining of cell wall is desirable. Ruthenium red was used for this purpose.

The meiotic division in the pollen mother-cells occurs in late April. The smear method was used for the study of these cells. In the first division metaphase 11 bivalent chromosomes were clearly counted (Text-fig. 2, a). Miss ROBERTSON studied the meiosis in the pollen mother-cells of *T. californica*, but owing to bad fixation she was not able to estimate the number



Text-fig. 1. Mitosis in endosperm. a, polar view. b, side view. $\times 2200$.

of chromosomes. She made also an attempt to count the chromosomes in the dividing endosperm nuclei and came to the conclusion that probably 8 was the haploid chromosome number of *T. californica*. The present writer was, however, able to count as many as eleven chromosomes in a dividing endosperm nucleus of *T. nucifera* (Text-fig. 1). One among the chromosomes of this plant has a distinct secondary constriction. It is remarkable that both *Taxus* and *Cephalotaxus* have 12 chromosomes in a haploid generation (ISHIKAWA, 1916; SAX, 1933; SUGIHARA, 1940). In the germination of microspores no prothallial cells are produced (Text-fig. 2, b).



Text-fig. 2. a, 1st division metaphase of the meiosis in a microspore mother-cell. b, germination of a microspore. c, body-cell with stalk cell nucleus and tube cell nucleus. d, mitosis in a body-cell. e, two sperm nuclei in common cytoplasm of a body-cell. a, $\times 1650$ b-e, $\times 850$.

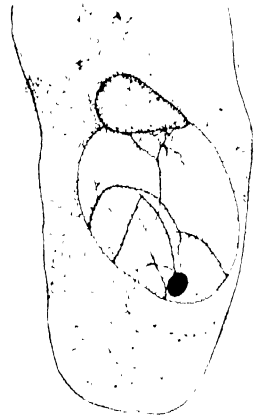
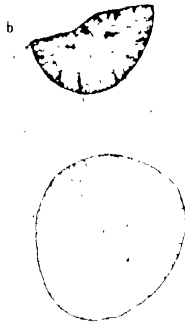
The tetrad division of the megaspore mother-cell occurs at the beginning of June. The endosperm formation is normal. Long tube-like cells radiating from the centre are observed.

Fertilization takes place in the middle of August. Just before fertili-

zation the nucleus of the body-cell, which is found at this time near the end of a pollen tube, divides (Text-fig. 2, d). The nucleus of the body-cell is seen always in the upper extremity of the cell. In contact with the lower extremity of the body-cell, a tube nucleus and a stalk-cell nucleus are observed (Text-fig. 2, c). The two sperm nuclei resulting from the division of the body-cell nucleus are very unequal in size, the smaller one being situated in the upper extremity. Two sperm nuclei are found embedded in common cytoplasm (Text-fig. 2, e). In this respect this plant agrees with *T. californica*. In *T. taxifolia*, according to COULTER and LAND a separate mass of cytoplasm surrounding two male nuclei is formed.



Text-fig. 3. a, mitosis in an archegonium to form ventral canal nucleus and egg nucleus. b, ventral canal nucleus and egg nucleus. $\times 850$.



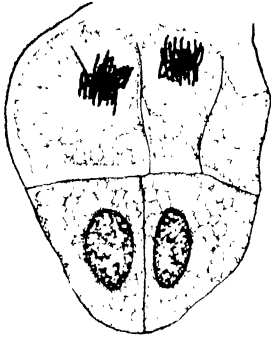
Text-fig. 4. Fertilization. $\times 170$.

The male nucleus is much smaller than the egg nucleus (Text-fig. 4). Dense cytoplasm is seen around the upper portion of the fusion nucleus (Pl. XVII, Fig. 1). As Miss ROBERTSON says, this was probably brought by the male nucleus.

The number of archegonia in each ovule is variable, three being most common. In this respect *T. nucifera* resembles *T. californica*. COULTER and LAND report that in *T. taxifolia* a single archegonium is seen in each ovule. Before fertilization, division of the central nucleus of the archegonium occurs to form ventral canal nucleus and egg nucleus. In this division 11 chromosomes were counted (Text-fig. 3). The ventral canal nucleus disintegrates soon after its formation. Early writers could not see the actual existence of this nucleus in the other species of *Torreya*.

Embryonal development soon follows the fertilization. At first 3

simultaneous nuclear divisions take place to form 8 nuclei. Cell wall formation begins in the 8 nucleus stage (Pl. XVII, Figs. 2-6). It is noteworthy that in *T. taxifolia* and *T. californica* the initiation of the cell wall is seen in the four nucleus stage.



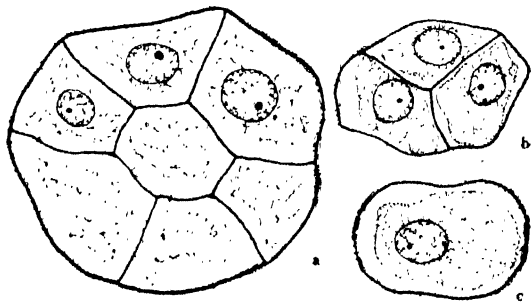
Text-fig. 5. Mitosis in the upper tier of a proembryo to cut off rosette nuclei. $\times 470$.

In *T. nucifera* the cells of a proembryo are arranged at first in two tiers. The number of cells in each tier is indeterminate. Before elongation of the suspensor, the cells of the lower tier may subdivide to form embryonic initials, the number of which is usually four. In the cells of the upper tier a wall is not formed on the side towards the general cytoplasm of the archegonium. COULTER and LAND emphasize the complete filling of the egg by the proembryo. But this phenomenon is not common to all species of *Torreya*; at least in *T. nucifera* the formation of the pro-

embryo is carried out in the basal portion of the egg cell. The nuclei descend to the bottom of the egg in the 4 nucleus stage (Pl. XVII, Fig. 4).

In the next stage nuclear division occurs in the cells of the upper tier of the proembryo to cut off the rosette nuclei (Text-fig. 5) and this gives rise to a proembryo consisting of three tiers (Pl. XVII, Fig. 7). The cells of the middle tier form the prosuspensor, which later elongates to enormous length. During the winter, development comes to a rest. And at the end of May of the next year the prosuspensor cells begin to elongate again. An embryo in this stage is shown in Pl. XVIII, Fig. 8. Cross sections through different zones of such an embryo are figured in Text-fig. 6.

In early June of the second year, divisions of the embryonic initials begin. The development of these cells goes on separately and gives rise to independent embryos (Pl. XVIII, Figs. 9-14). The first division of the embryonic initials seems to be longitudinal. Then



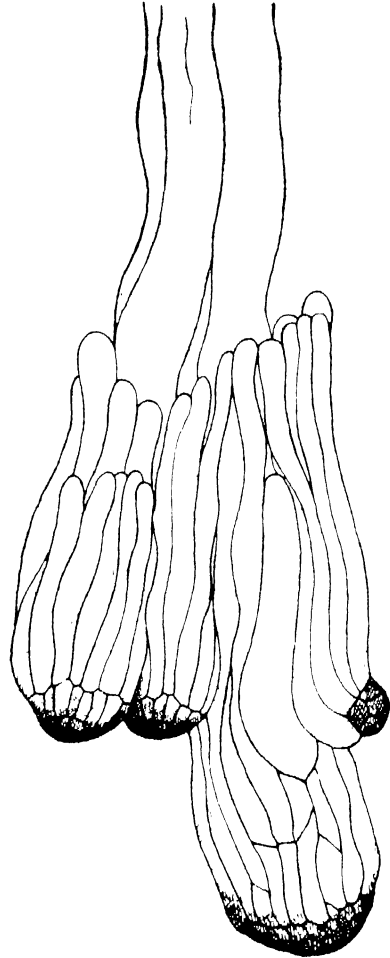
Text-fig. 6. Serial cross sections through different zones of an embryo in the second year growth. $\times 470$.

divisions of different directions follow one after another. A primary suspensor is not formed in this plant. Only numerous embryonal tubes are observed (Text-fig. 7). Thus *T. nucifera* is characterized by a significant cleavage polyembryony. Whether the same state of things is or is not common to other species of *Torreya* can not be determined, no definite description concerning this point being available at present. The occurrence of cleavage polyembryony in *Torreya* is rather astonishing, because this phenomenon is not observed in *Taxus* and *Cephalotaxus*.

In *Torreya* embryonal development proceeds keeping pace with the development of endosperm. So the tortuous twisting of elongating suspensor cells, which is very common in Coniferales, is not observed in early stages of the embryonal development.

COULTER and LAND state in the paper above cited, "In several instances a number of small embryos were observed embedded in endosperm about the suspensor region of the normal embryo. Our material did not permit any determination of their origin,". In the present writer's opinion some of them would be rosette embryos. As shown in figures in Pl. XVIII, the development of rosette embryo is very common in *T. nucifera*. From the data above described *Torreya nucifera* appears to be the most primitive species of *Torreya*.

In several cases, sometimes before cell wall formation begins, the present writer was able to count diploid chromosome number (22) in



Text-fig. 7. Four distinct embryos attached at the end of a prosuspensor. $\times 90$.

endosperm cells (Pl. XVIII, Fig. 15). In what manner such a cell has arisen cannot be determined at present.

SUMMARY

1. Meiotic division in the microspore mother-cells occurs at the end of April. The haploid chromosome number of this plant is estimated to be eleven.

2. Meiotic division in the megaspore mother-cell takes place at the beginning of July. In the endosperm formation long tube-like cells radiating from the centre are formed.

3. In the germination of the microspore no prothallial cells are observed. The nucleus of the body-cell divides just before fertilization. At this time the nucleus is situated at the upper extremity of the cell. The two sperm nuclei are very unequal in size, the smaller one being found in the upper extremity of the body-cell. Two sperm nuclei are found in common cytoplasm.

4. Fertilization occurs in the middle of August. Dense cytoplasm is found around the upper portion of the fusion nucleus.

5. After fertilization 3 simultaneous nuclear divisions are carried out. Cell wall formation begins at the 8 nucleus stage. The proembryo is formed in the basal part of the egg cell.

6. Usually four embryonic initials are formed at the end of the pro-suspensor. These embryonic initials develop independently into 4 separate embryos. *Torreya nucifera* is thus characterized by the cleavage polyembryony. In the development of the embryo a primary suspensor is not formed. Only numerous embryonal tubes are observed. Rosette embryo is very common. *T. nucifera* perhaps may be the most primitive species of *Torreya*.

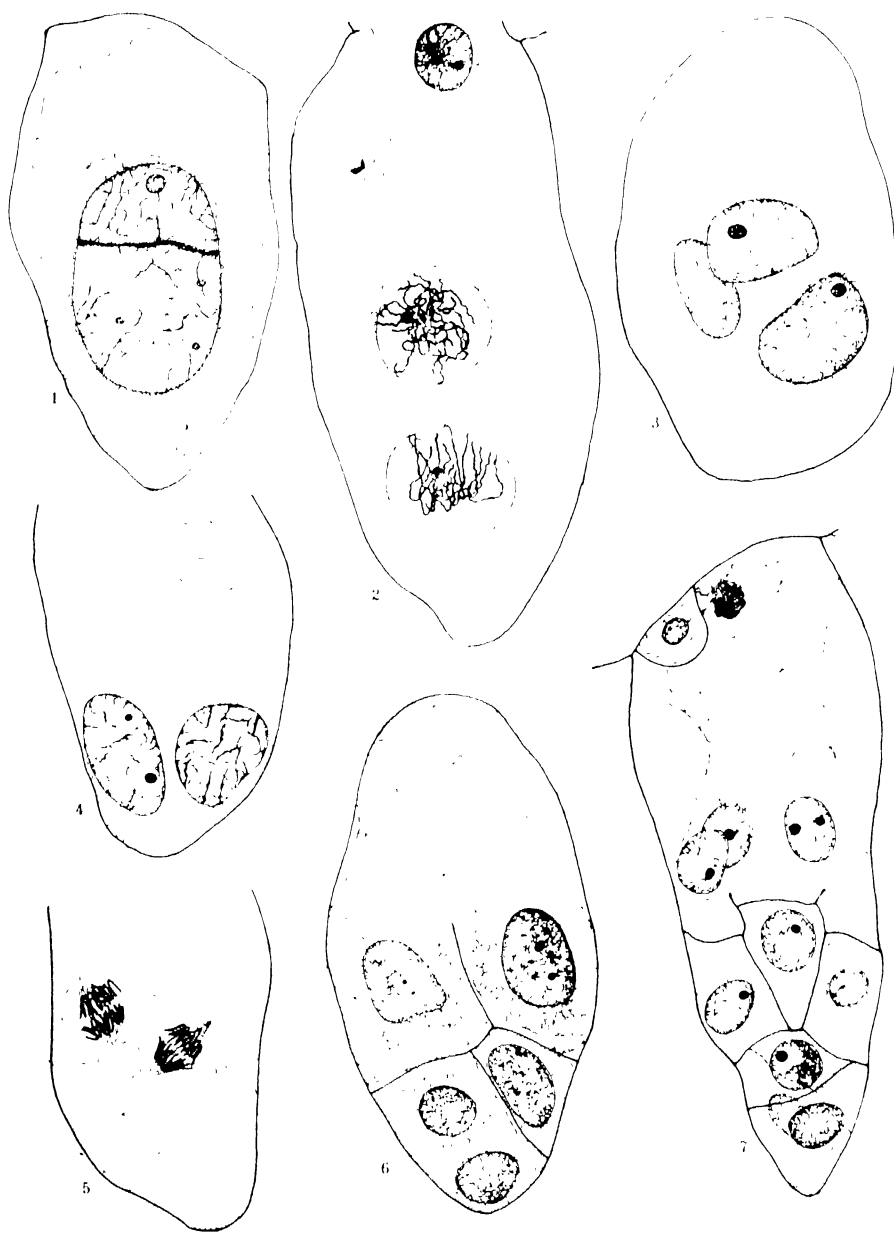
LITERATURE

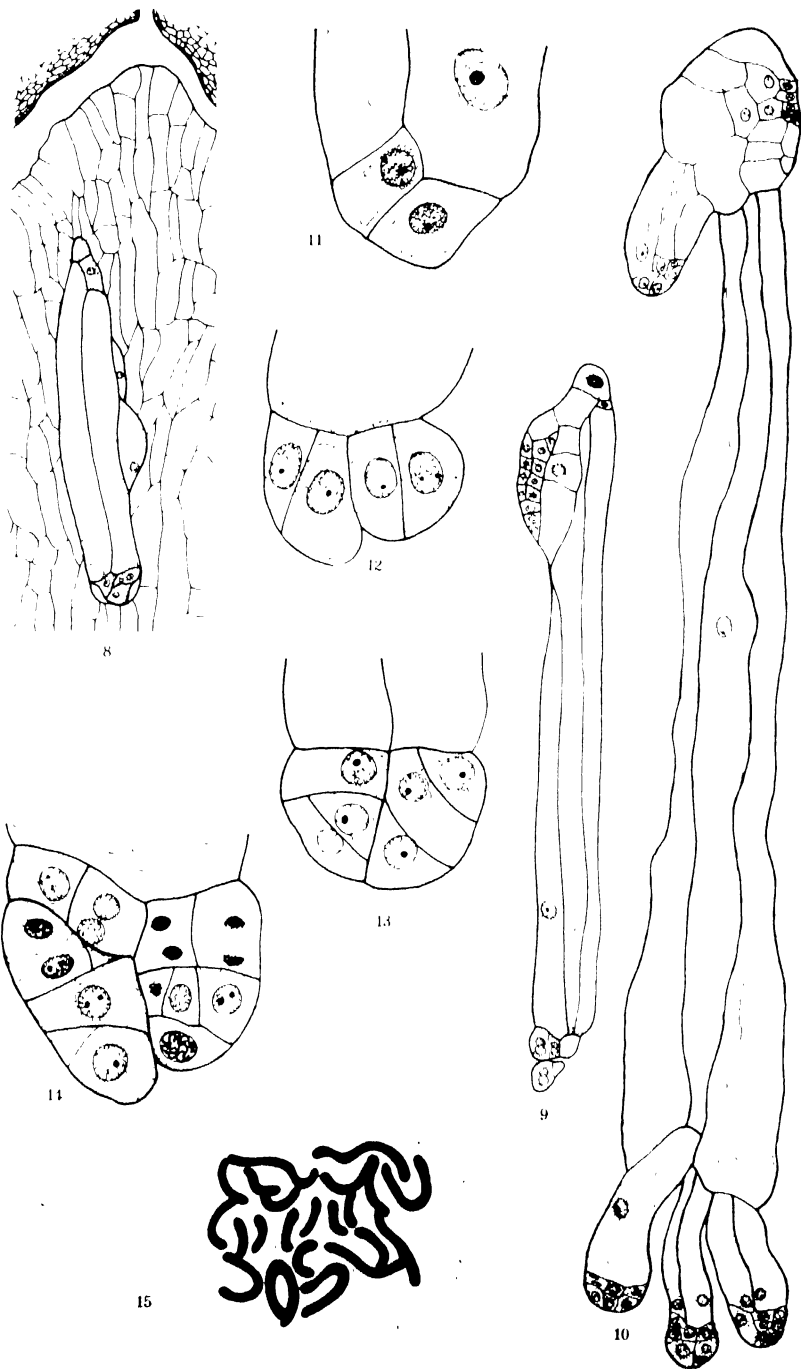
- BUCHHOLZ, JONE T. 1929. -- The Embryogeny of the Conifers. Proc. Intern. Cong. Plant Sci.
- COKER, W. C. 1907. -- Fertilization and Embryogeny in *Cephalotaxus Fortunei*. Bot. Gaz. 43.
- COULTER, JONE M. and LAND, W. J. G. 1905. Gametophyte and Embryo of *Torreya taxifolia*. Bot. Gaz. 39.
- ISHIKAWA, M. 1916. -- A List of the Number of Chromosomes. Bot. Mag. Tokyo. 30.
- JÄGER, L. 1899. -- Beiträge zur Kenntniss der Endosperm-bildung und zur Embryologie von *Taxus baccata*. Flora. 86.

- ROBERTSON, AGNES. 1904. - Studies in the Morphology of *Torreya californica* TORREY. *New Phyt.* **3**.
- SAX, KARL and SAX, HALLY JOLIVETTE. 1933. - Chromosome Number and Morphology in the Conifers. *Jour. Arnold Arb.* **14**.
- SUGIHARA, Y. 1940. - On the Multipartite Chromosome-ring in *Cephalotaxus drupacea* SIEB. et ZUCC. *Sci. Rep. Tōhoku Imp.* **15**.

EXPLANATION OF PLATES XVII AND XVIII

Fig. 1. Fusion nucleus. Fig. 2. Two nucleus stage of a proembryo. Figs. 3. and 4. Four nucleus stage. Fig. 5. Third division to form 8 nuclei. Fig. 6. Eight nucleus stage. Fig. 7. Still later stage. Figs. 8-10. Embryo development in the second year. Figs. 11-14. Independent development of embryonic initials. Fig. 15. Diploid nuclear plate in an endosperm cell. Magnification:— Figs. 1-7, $\times 470$. Figs. 8-10, $\times 90$. Figs. 11-14, $\times 360$. Fig. 15, $\times 1650$.





AN ANATOMICAL STUDY OF *TUBIFEX* (*PELOSCOLEX*)
NOMURAI, N. SP., OBTAINED AT THE BOTTOM OF
LAKE TAZAWA, THE DEEPEST LAKE OF JAPAN

By

GOTARÔ YAMAMOTO

*Aomori Fishery Laboratory, Institute of Agricultural Research,
Tôhoku Imperial University, Aomori*

AND

KATSUHIRO OKADA

Biological Institute, Tôhoku Imperial University, Sendai

(With 20 text-figures)

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SYNOPSIS

Tubifex (*Pelosclex*) *nomurai* is a new aquatic oligochaete, obtained at the depth of about 425 m. at the bottom of Lake Tazawa, one of the deepest lakes in Japan. A diagnostic description and the result of an anatomical investigation of *Tubifex* (*Pelosclex*) *nomurai* are given in this paper. And general considerations are briefly stated concerning the *Tubifex*-group of the Family Tubificidae.

INTRODUCTION

In the present paper, the writers deal with an anatomical study of a newly discovered aquatic oligochaete, obtained at the bottom of Lake Tazawa, the deepest lake of Japan. This freshwater lake is situated at the southwestern foot of Mt. Komagadake in the Ôu mountain range in the Senboku District, Akita Prefecture. It is a round caldera lake surrounded by a range of low hills. The surface of the lake is 250 m. above the sea-level, 19.5 km. in circumference and 25.65 km². in area. The deepest point of this lake is a little southwest of the centre, where the depth measures 425 m., just 175 m. below the sea-level. The water of this lake is a beautiful indigo in colour, approaching nearly the Forelian Colour No. 2 of standard colours. In degree of transparency this water is usually graded as being in the second place among the lakes of Japan, and in the fourth place among the lake-waters of the whole world.

Several studies have been made of the aquatic oligochaetes, obtained from the deep lakes of the world. W. MICHAELSEN ('02, '03, '05 & '26)

explored not only the lakes of Germany, but also Lake Baikal in Siberia. W. B. BENHAM ('03 a & b) explored every lake of New-Zealand. F. E. BEDDARD ('06) explored Lake Tanganyika in Africa. Besides these scientists, S. HRABÈ ('31) carefully explored Lake Ochrida in the Balkan Peninsula, but, in Japan no report had been made of the aquatic oligochaetes of the deep lakes, in spite of the abundance of such lakes, viz., Lake Sikotu, Hokkaido, and Lakes Tazawa and Towada, Akita Prefecture. Prof. Dr. E. NOMURA has long been interested in exploring these deep lakes. He first went to Lake Tazawa with Mr. J. ÔIZUMI and obtained two immature specimens of the species in the summer of 1935. Mr. J. ÔIZUMI went there alone again in the autumn of 1935 and succeeded in obtaining many more immature specimens. Further, Mr. I. HAMAI went there alone in June of 1937 and succeeded in obtaining many mature specimens.

The present investigation was undertaken in April of 1939 at the suggestion of Prof. E. NOMURA. The species under investigation is closely allied to the species of *Tubifex*-group described already, but is not identical with them. Therefore, the writers give a diagnostic description of the present species in the following pages and beg to propose the new name, *Tubifex* (*Pelosclex*) *nomurai*. This specific name is dedicated in honour of Prof. E. NOMURA, as being the first discoverer of the species.

Before proceeding further, the writers wish to express their hearty thanks to Prof. E. NOMURA for his friendly guidance and to Dr. I. MOTOMURA for his valuable advice during this investigation. The writers are, also, indebted to Mr. I. HAMAI for the kind loan of his specimens, to Dr. S. ÔHFUCHI for the free inspection to his library, and to Mr. E. HIRAI for his helpful friendship in taking photomicrographs.

DESCRIPTION OF *TUBIFEX* (*PELOSCOLEX*) *NOMURAI*

EXTERNAL FEATURES (Fig. 1): - Size of fully grown specimen 7-8 cm. long in whole length and 1-1.2 mm. thick in clitellum, the thickest portion of body. Number of segments commonly 90-120 and occasionally though rarely 140. Clitellum forming a nearly complete ring, occupying from posterior two-thirds of Segment X to Segment XII. Colour of body-wall (except clitellum) in live specimen pinkish-grey and somewhat transparent. Colour of clitellum creamy-white and opaque. Prostomium prolobous and not conspicuous. Each pre-clitellar segment small. Pre-clitellar portion

of body only 3 mm. long. Each post-clitellar segment (except terminal portion) considerably larger. In fully mature specimen, cuticular papillae measuring 10–15 μ in height, present in clitellar portion, especially dense around male pores. Dorsal seta-bundles lying on both dorso-lateral corners of body, and ventral seta-bundles on both ventro-lateral corners. Setae, both dorsal and ventral, commence in Segment II. Dorsal setae capillary and pectinate, lying to fairly posterior portion of body. Pectinate seta stretches thin membrane between the two terminal prongs. Ventral setae bifurcate and slender. No ventral seta in fully mature specimen present posterior to Segment IX. No penial and genital setae present.

INTERNAL CHARACTERS: Mouth in Segment I and buccal cavity reaches to the anterior end of Segment II. Pharynx in Segments II and III, covered mainly with pharyngeal gland. Oesophagus in Segments IV–VI and in the anterior two-thirds of Segment VII. Oesophageal gland in Segment IV. Chloragogue coat commences in Segment VI and intestine in the posterior one-third of Segment VII. Brain consists of a median and two lateral lobes. Each lateral lobe producing anterior larger and posterior smaller branches. Anterior nephridia paired in Segments VII and VIII. Posterior nephridia commence in Segment XIV, paired. Two pairs of intestinal hearts connect supra-intestinal vessel to ventral vessel in Segments VIII and IX. Intestinal network of blood-vessel well-developed.

Genital organs typical in Tubificidae (Fig. 2). Testes paired in Segment X. Sperm-sacs consist of anterior and posterior ones, present along the median dorsal line of alimentary tract. Anterior sperm-sac, being anterior diverticulum of Septum IX/X, reaches to the anterior end of Segment IX. Posterior sperm-sac, being posterior diverticulum of Septum



Fig. 1. Photograph, showing external feature of type specimen of *Tubifex (Peloscolex) nomurai* viewed from dorsal side. $\times 1.8$

X XI, reaches to the posterior end of Segment XIII. Sperm-ducts paired, each consists of sperm-duct funnel, vas deferens, and atrium. Each sperm-duct funnel ciliated, opens widely, attaching the anterior face of Septum X XI. Each vas deferens convoluted and continuous posteriorly to atrium, measuring about two or more times as long as atrium. Each atrium thick-walled, consists of anterior part, wider in diameter, and posterior part, narrower in diameter. Large prostate gland opens to the middle of wider part of atrium. Penis protrusile, without chitinous sheath. Paired penial chambers open to the posterior one-fourth of Segment XI on ventral setal lines. Ovaries in Segment XI, paired. Ovisac, being posterior diverticulum of Septum XI XII, reaches to the middle of Segment XIV. Oviducts paired, each oviduct consists of oviduct funnel and duct proper. Each oviduct funnel attaching the anterior face of Septum XI XII, ciliated and continuous to narrow duct proper. Each oviduct proper short, lying along posterior face of Septum XI XII. Openings of oviducts present at inter-segmental furrow between Segments XI and XII

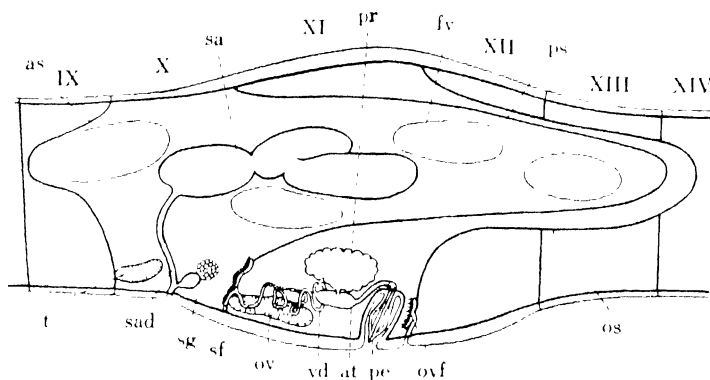


Fig. 2. Schematic representation of genital organs of *Tubifex Peloscolex nomurai*, viewed from left side. $\times 30$. IX-XIV No. of segment; as anterior sperm-sac, at atrium of sperm-duct, fv floating vesicle, os ovisac, ov ovary, ovf oviduct funnel, pr penis, pr prostate gland, ps posterior sperm-sac, sa spermathecal ampulla, sad spermathecal duct, sf sperm-duct funnel, sg spermathecal gland, t testis, vd vas deferens.

on ventral setal lines. Spermathecae paired, each spermatheca consists of ampullar portion and duct with gland. Openings of spermathecal ducts present at the middle of Segment X. Small thin-walled sacs, named floating vesicles by ourselves, present within both anterior and posterior sperm-sacs, counting 7-9 in number. Posterior end of each floating vesicle

having supposed duct, slender and continuous to the interior of sperm-sac.

HABITAT (Fig. 3): Oozy bottom of Lake Tazawa, Akita Prefecture.



Fig. 3. Lake Tazawa, viewed from northeastern shore.

MATERIAL AND METHOD

The materials used in this investigation were the fixed specimens as well as the live ones. The fixed specimens were chiefly those collected by Mr. I. HAMAI in 1937. In order to supplement the observation, fresh specimens were collected in June and July of 1939 by G. YAMAMOTO, one of the present writers.* For collecting purposes a small dredge with a rope of 1000 m. was applied in the deepest part of the lake, where the specimens are always found most abundantly. And also from the same part of the lake many slender tubes of this animal were obtained, which were greenish-black in colour and constructed of smooth, thin walls of secreted mucous cohering with ooze. These tubes measured 6-7 cm. in whole length and mostly 1-1.5 mm., rarely 2 mm., in caliber, though they were usually broken up into a few pieces (Fig. 4).

After narcotisation in a dilute alcohol, the specimens were quickly fixed with Rath's solution without glacial acetic acid, and then preserved in a 80% solution of alcohol. For these hardened specimens, the sections were made serially, 10 μ or 15 μ in thickness by the paraffin method, and stained with Delafield's haematoxylin-eosin or by Mallory's connective tissue staining method.



Fig. 4. Photograph, showing tube of *Tubifex* (*Peloscolex* *nomurai*). $\times 1.8$.

*The writers' thanks are due to Mr. JUNZI YAMATE for his assistance in collecting the specimens.

ANATOMY AND HISTOLOGY

I. SEGMENTATION AND ANNULATION OF BODY

In the specimen sexually matured, the body measures about 7-8 cm. in length. The thickness of the body is about 1 mm. in most segments and is about 1.2 mm. in the clitellum. At the most anterior end of the body is the prostomium, which shows a probolous structure with a bluntly rounded anterior end. This prostomium is followed by the peristomium or Segment I, which is single and non-annular. The so-called secondary annulation of the body-segments is formed in Segments II-V of the pre-clitellar segments. Segment II consists of two annuli, the shorter anterior and the longer posterior annulus. In Segments III and IV, each consists of three annuli. Of these three annuli, the anterior two are the shortest, nearly equal in length, the posterior one being a little longer. In Segment V, four annuli are observable. The anterior two and the most posterior one are shorter, all three being nearly equal in length, and the third one is a little longer. The remaining pre-clitellar body-segments are all single and non-annular. All these pre-clitellar segments are comparatively short. The total length of the prostomium and the pre-clitellar segments is no more than 3 mm.

The clitellum occupies from the posterior two-thirds of Segment X to the end of Segment XII, and forms a nearly complete ring. The post-clitellar body-segments arise posterior to Segment XII. Most of the post-clitellar segments are notably long, each of them being generally about 1 mm. The remaining post-clitellar segments become again short near the terminal portion of the body, which diminishes gradually in thickness posteriorly towards the last segment. The number of segments is from 90 to 120 in most cases, and occasionally though rarely as much as 140.

II. SETAE AND SETA-BUNDLES

Three kinds of the setae are found in this species. One kind is capillary. These capillary setae, without any sort of spine, measure about 0.5-0.6 mm. in length. The other two kinds of setae are bifurcate. Of these bifurcate setae, one has a thin membrane between the two prongs, and may be called a kind of pectinate seta. The pectinate setae are of an incomplete sigmoid type, no clear nodule being situated. In each pectinate seta, the two prongs directed upwards are sharply pointed, and are about equal to one another in stoutness. A few stripes but no special

notch are observable in the thin membrane between the two prongs, like that of *Tubifex ignotus* and of *Tubifex barbatus*. The length of the pectinate setae is discordant, and gradually decreases posteriorly (Fig. 5 A). Generally speaking, they are one-fourth or one-fifth as long as the capillary setae. Another kind of bifurcate setae is of the so-called sigmoid type, the nodule being situated at about the middle of the setal length. In each sigmoid seta, the upper prong is naturally longer than the lower. It is apparent, however, that the difference in the length of the two prongs becomes less noticeable in those of the posterior segments (Fig. 5 B). The size of the sigmoid setae is a little larger than that of the pectinate ones.

In each chaetigerous segment, four seta-bundles, the two dorsal and the two ventral, are arranged in the characteristic manner of the Family Tubificidae. The paired dorsal seta-bundles composed invariably of the

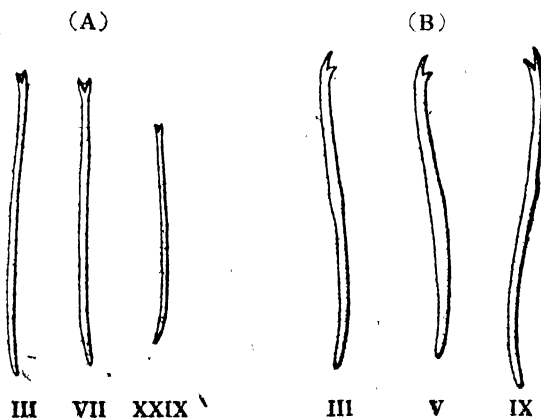


TABLE 1. One example showing number of setae contained in each dorsal seta-bundle of *Tubifex (Peloscolex) nomurai*, and two other examples showing number of setae contained in each ventral seta-bundle respectively.

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capillary and the pectinate setae, are situated at both the dorso-lateral corners of each single segment, and the paired ventral seta-bundles composed of the bifurcate sigmoid setae, are situated at both the ventro-lateral corners of the same. In secondarily annulated segments, each of the longest annuli bears these seta-bundles. The number of the setae in each seta-bundle decreases posteriorly (Table 1).

III. BODY-WALL, SEPTA, AND SEPTAL SACS

THE BODY-WALL.

The body-wall of the species is generally thin with the exception of the clitellum and the antero-terminal portion of body.

The Cuticular Layer. This outermost layer of the body-wall is well-developed in the pre-clitellar portion, and becomes thinner in the clitellum and the post-clitellar portion. Irregularly shaped blackish pigment granules are scattered annularly in the cuticular layer of the pre-clitellar portion. Such distribution of the pigments is one of the characteristics of this species. This pigmented cuticle is thickened notably and measures 20–25 μ in thickness (Fig. 6). There are, however, no sensory papillae in this pigmented cuticle, and also there is no sensory cell in the hypodermis in connexion with the pigmentation. In the sexually matured specimen, special papillae are formed and arranged irregularly in the cuticular layer of the clitellum, especially abundant around the male pores. These papillae, in which fine stripes may or may not be present longitudinally, are simple in structure and are nothing but the cuticular processes with round or flattened ends (Fig. 7). The number of these papillae of this species is also far less than that of the other species of *Peloscoides*.

The Hypodermis. The extra-clitellar hypodermis is less than 6 μ in thickness, thicker in the pre-clitellar portion and thinner in the post-clitellar portion, and is a unicellularly arranged layer of the hypodermal cells. The nucleus of each hypodermal cell is nearly spherical and is rich in chromatin granules, deeply staining with Delafield's haematoxylin. Vacuoles and small masses of fine granules are often present in its cytoplasm. The clitellar hypodermis is far thicker than the extra-clitellar one, and measures about 30 μ in thickness. In cross sections through the clitellum, the outline of the body shows a rounded square, owing to the well-developed hypodermis at the dorso-lateral and ventro-lateral corners of both sides (Fig. 8). The clitellar hypodermis is, also, a unicellular layer, abundantly intercalated with columnar gland cells and sup-

ported by the supporting cells. The cytoplasm of this gland cell is rich in granules, deeply staining with Delafield's haematoxylin.

The Circular and Longitudinal Muscle Layers. These two muscle layers are not well-developed in this species, like those of *Tubifex tubifex* or *Tubifex hattai*.

The Peritoneal Layer. The peritoneum is only a thin lining inside the integument of the species and is far less developed than that of *Tubifex hattai*. The lateral lines are easily detected in the pre-clitellar portion. They commence in the middle of Segment III and reach to the middle of Segment VIII without cessation. In Segments V and VI, they are quite distinct from the peritoneum by their peculiarly cellular structure. In Segments III, IV, VII and VIII, they are closely attached processes with the peritoneum, and push the longitudinal muscle fibres aside (Fig. 6).

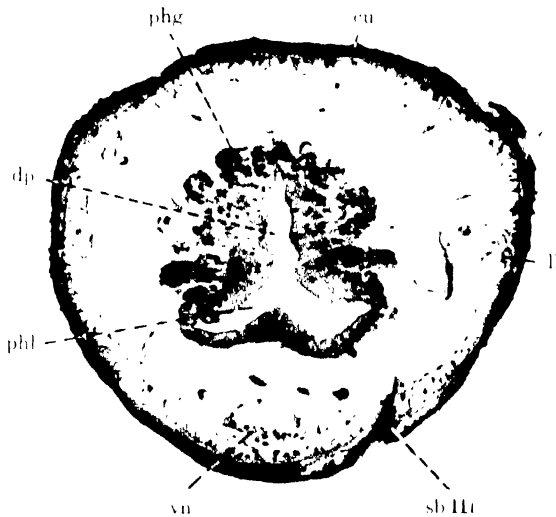


Fig. 6. Photomicrograph of transverse section through pharyngeal portion. $\times 95$. *cu* cuticle, *dp* dorsal pouch of pharynx, *ll* lateral line, *phg* pharyngeal gland, *phl* lumen of pharynx, *sb III* seta-bundle of Segment III, *vn* ventral nerve cord.

THE SEPTA.

The septa begin at Intersegment III IV. In this species, they are mostly set obliquely in a constant angle round the intestine, the anterior end being situated in the dorsal side and the posterior end in the ventral side, while, in many species of Tubificidae described already, the septa of

the pre-clitellar segments are funnel-shaped with the smaller end directed posteriorly. The musculature of the septa are most distinct around the intestine.

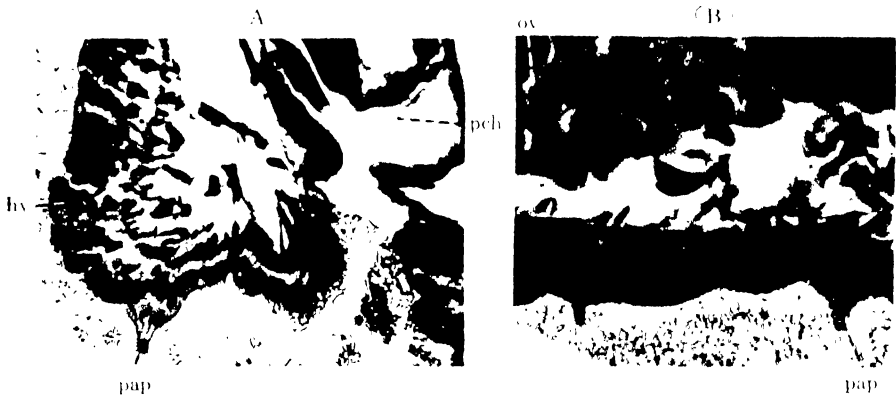


Fig. 7. Two photomicrographs, showing cuticular papillae. (A) Transverse section through clitellum. $\times 550$. (B) Longitudinal section through clitellum. $\times 380$. *hy* hypodermis, *ov* ovary, *pap* papillae, *pch* penial chamber

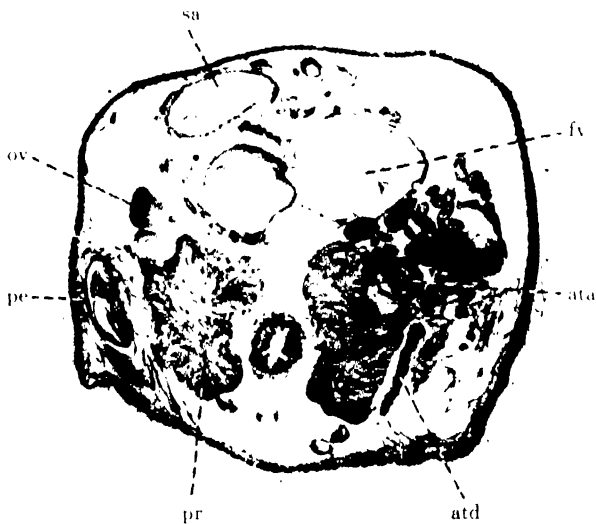


Fig. 8. Photomicrograph of transverse section through clitellum. $\times 50$. *ata* ampullar portion of atrium, *atd* duct portion of atrium, *fV* floating vesicle, *ov* ovary, *pe* penis, *pr* prostate gland, *sa* spermathecal ampulla.

THE SEPTAL SACS.

Two pairs of septal sacs are found only in the fully mature specimens. These sacs are attached to the posterior faces of Septa V/VI and VI/VII on both sides of the ventral vessel which extends its branches inside the sacs. The wall of each septal sac is a structureless thin membrane. As reported already by E. NOMURA (13) in the case of *Limnodrilus gotoi*, the anterior end of the septal sac is also adjacent to the chloragogue cells in the case of *Tubifex (Peloscolex) nomurai*. In each septal sac, many cells resembling the chloragogue cells and a few tubules in a convoluted situation are observed together with the above-mentioned branches of the ventral vessel (Fig. 9). The

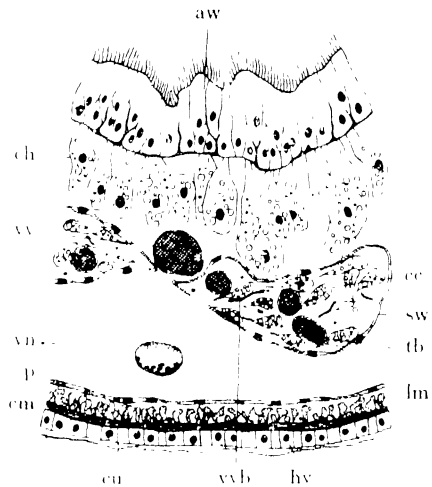


Fig. 9. Schematic drawing of transverse section through Segment VI, illustrating structure of septal sac. 350. *aw* wall of alimentary tract, *ce* cells found in septal sac, *ch* chloragogue cell, *cm* circular muscle, *cu* cuticular layer, *hp* hypodermis, *lm* longitudinal muscle, *p* peritoneum, *sw* wall of septal sac, *th* convoluted tubule found in septal sac, *vn* ventral nerve cord, *vxb* branch of ventral blood vessel, *vv* ventral blood vessel.

lumen of the convoluted tubule inside the sac is filled with yellowish substances, which may be identical with a metamorphosed fat-body often found in various organs of many oligochaetes.

IV. ALIMENTARY SYSTEM

The Mouth and the Buccal Cavity (Fig. 10). The mouth opens on the antero-ventral end of Segment I, and the buccal cavity reaches to the beginning of Segment II. The wall of the buccal cavity is homologous to the body-wall in structure: the innermost cuticular layer is non-ciliated and contains a little blackish pigment, and the hypodermal layer, which is constituted of unicellularly arranged columnar cells, is well-developed, though the cuticle is thinner and the muscle layers are less-developed than those of the body-wall. The posterior end of the buccal cavity is in direct continuation to the pharynx.

The Pharynx and the Pharyngeal Glands (Fig. 6). The pharynx lies

in Segments II and III. The lumen of the pharynx shows a Λ -shape, in cross sections, owing to the formation of its dorsal pouch, and is lined with endodermal columnar cells forming a ciliated epithelium, which is clearly distinguished from the wall of the buccal cavity. The outer side of the pharyngeal wall is covered with the thin connective tissue derived from the peritoneum. Some muscle fibres are found at this outer layer. The ventral wall of the pharynx is thinner anteriorly and thicker posteriorly. The cytoplasm of the endodermal cells of the pharyngeal wall shows numerous fine strands running perpendicularly to the wall. The pharyngeal glands are small cell-masses attached densely to the dorsal and lateral faces of the entire pharyngeal wall. Each cell-mass is composed of pear-shaped gland cells; their proximal ends are prolonged into small ducts. In each cell-mass, these ducts unite with each other and form a common duct, which opens to the pharyngeal cavity through the interspaces of the endodermal cells.

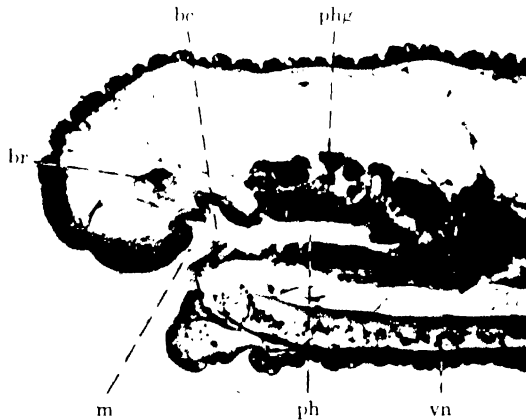


Fig. 10. Photomicrograph, showing anterior end of alimentary tract in about median sagittal section through anterior terminal portion of body. $\times 120$. *bc* buccal cavity, *br* brain, *m* mouth, *ph* pharynx, *phg* pharyngeal gland, *vn* ventral nerve cord.

The Oesophagus and its Appendages. The oesophagus occupies from the beginning of Segment IV to the anterior two-thirds of Segment VII. The oesophageal lumen shows, in cross sections, an elliptical shape flattened dorso-ventrally. By this peculiar shape of the lumen, the oesophagus is distinguished from the pharynx. The wall of the oesophagus constituted of columnar endodermal cells, forms also a ciliated epithelium, which is covered with the muscular connective tissue. The endodermal wall of

the oesophagus is rich in longitudinal foldings internally. Digestive glands are also found at the dorsal and lateral faces of the oesophageal wall in Segment IV. These glands may be called the oesophageal glands and show the same structure as the pharyngeal glands. Some chloragogue cells are developed in Segments VI and VII.

The Intestine and its Appendages (Figs. 11 & 12). The intestine begins at the posterior one-third of Segment VII. The outline of the beginning portion of the intestine is roundish-triangular in cross sections. By this peculiar shape, the intestine is distinguished clearly from the oesophagus. The outline of the intestine in Segment XII becomes round or elliptical in cross sections. The intestinal lumen becomes increasingly spacious at the posterior portion, while, the thickness of the intestinal wall decreases gradually from Segment XII to the last segment. In the portion posterior to Segment XX, the lumen measures 250-300 μ in caliber, and

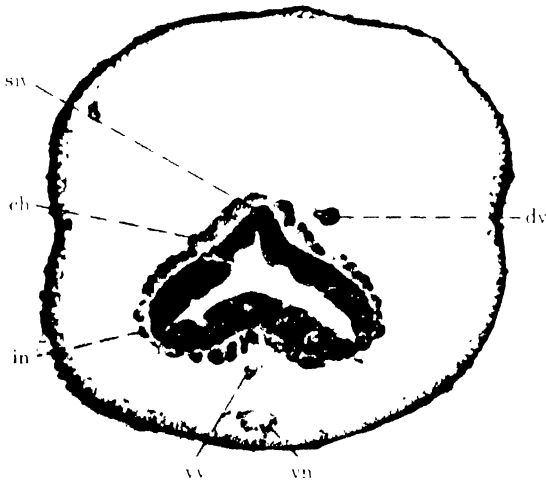


Fig. 11 Photomicrograph, showing beginning portion of intestine in transverse section. $\times 85$ ch chloragogue coat, dv dorsal vessel, in intestine, siv supra-intestinal vessel, vn ventral nerve cord, vv ventral vessel.



Fig. 12 Photomicrograph, showing posterior end of body in transverse section. $\times 110$ al anal lip.

the wall 30-50 μ in thickness. The terminal end of the intestine extrudes slightly outside the body-wall and then opens to the anus, the anal lip resulting there. The body-wall of the last segment covers this lip from the dorsal and lateral sides. The intestinal wall consists of two layers, the inner endodermal epithelium and the outer thin peritoneum. This endodermal epithelium is formed by unicellularly arranged columnar cells. The

inner surface of this epithelium is furnished with the secreted cuticle and the rather long cilia in comparison with those of the pharyngeal and oesophageal epithelia. The nucleus of each endodermal cell is ellipsoidal and takes a nearly central position of the cell, one distinct nucleolus always appearing. The intestinal network of blood vessels is well-developed in the intestinal wall of Segments VII-XX. In this portion, the capillary blood vessels are intercalated into interspaces of the endodermal cells. The chloragogue coat begins at Segment VI as stated above, and is well-developed around the intestinal wall of Segments VII-XXXV. In



Fig. 13. Photomicrograph, showing paired structure of posterior nephridia. $\times 150$. *ch* chloragogue coat, *in* intestine, *mb* intestinal network of blood vessel, *pn* posterior nephridia, *vn* ventral nerve cord.

more posterior segments to XXXV, the coat becomes gradually indistinct towards the terminal end of the intestine. The chloragogue coat is 30-45 μ in maximum thickness. The typical chloragogue cell is club-shaped with the thicker end directed towards the body-cavity, as described already by some authorities. The nucleus of the chloragogue cell lies always in the distal half of the cell and contains invariably greenish chloragosomes, which take no stain with Delafield's haematoxylin-eosin or by Mallory's connective tissue staining method. The large and the small type of the chloragosomes are always found

in each chloragogue cell, as stated by DIXON ('15) in the case of *Tubifex tubifex* and NOMURA ('13) in the case of *Limnodrilus gotoi*. Moreover, various transitory types of them between the large and the small ones are also found in the case of *Tubifex (Peloscolex) nomurai*. The cell-membrane of the chloragogue cell is structureless and is stained distinctly with the aniline blue of Mallory's staining method.

V. EXCRETORY SYSTEM

The Anterior (or Per-clitellar) Nephridia. A pair of the nephridia is found in Segment VII and VIII respectively. Each anterior nephridium consists of a ciliated funnel, a long convoluted tubule, an ampullar portion of duct and a duct proper in the order from the anterior to the posterior.

The ciliated funnel arises from the front of the anterior septum of the said segment, and is continuous to the tubule measuring $3-5\mu$ in diameter. This tubule passes through the anterior septum and enters the ventrolateral coelom of the said segment, where it shows an extensive convolution and then posteriorly continues to the duct. The anterior half of the duct forms the ampullar portion, measuring about 15μ in caliber and $40-50\mu$ in length, and the terminal half is the duct proper which becomes narrow again. The nephropore opens outside each ventral setal line of posterior level of the said segment.

The Posterior (or Post-clitellar) Nephridia. A pair of the posterior nephridia is found in the respective post-clitellar segment, and the first pair of them begins at Segment XIV. According to the descriptions reported already in most species of Tubificidae, the posterior nephridia of one side of the body are poorly developed and the paired formation of them is generally spoiled to some extent. In the present species, however, the posterior nephridia are equally well-developed on both sides of the body and the paired formation of them is typically expressed as in the case of the anterior nephridia (Fig. 13). The structure of each posterior nephridium is almost the same as that of the anterior one, although the convolution of the tubule and of the duct proper is more extensive in this case.

VI. NERVOUS SYSTEM

The Brain and its Nerve Branches (Fig. 14). The brain consists of a central lobe and both lateral lobes, and lies in the prostomium and Segment I, the posterior end of the brain often reaching to Segment II. The central lobe is an anteriorly swollen mass on the dorsal side of the buccal cavity and no nerve branch is found there. The lateral lobes are directly adjacent to both latero-ventral faces of the central lobe. Three nerves are branching from the lateral lobe on either side of the brain. The lateral prostomial nerve runs out forwards from the anterior border of the corresponding lateral lobe to the prostomial wall. The parietal nerve is somewhat small and runs out backwards from the postero-dorsal end of the corresponding lateral lobe to the dorsal body-wall. The peripharyngeal nerve runs out from the postero-ventral border of the corresponding lateral lobe and proceeds downwards along the curvature of the body-wall to reach the median wall of Segment II, where the peripharyngeal nerve unites with its fellow of the opposite side and then

continues to the anterior end of the ventral nerve cord. The peripharyngeal nerve is constituted mainly of nerve fibres and of little nerve cells.

The Ventral Nerve Cord and its Nerve Branches (Figs. 14 & 15). The ventral nerve cord begins at Segment II. In Segments II and III, the ventral cord does not give off as branches any lateral nerve but forms a segmental ganglion in each segment. This segmental ganglion lies near the posterior septum of the respective segment. In Segments IV and V, the ventral cord gives off as branches one pair of lateral nerves and

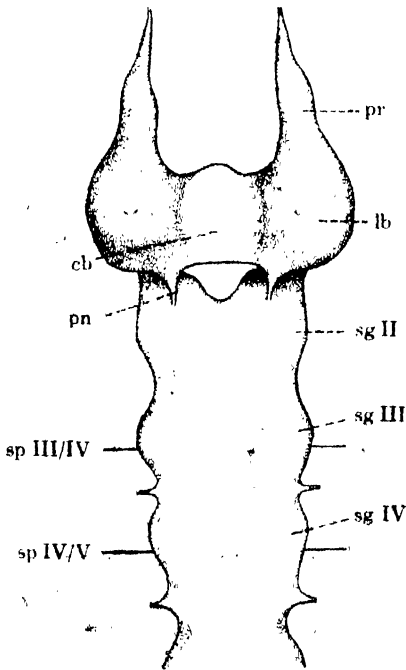


Fig. 14. Schematic representation viewed from dorsal side, showing brain and anterior part of ventral nerve cord. $\times 200$. *cb* central lobe of brain, *lb* lateral lobe of brain, *pn* parietal nerve, *pr* lateral prothoracic nerve, *sg* segmental ganglion, *sp* septum.

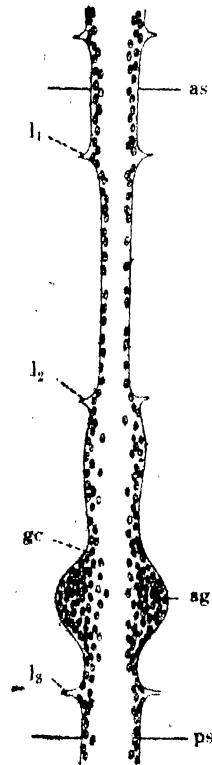


Fig. 15. Schematic representation viewed from dorsal side, showing ventral nerve cord in Segment VI. $\times 200$. *as* anterior septum, *gc* ganglionic cell, *l1* first lateral nerve branch, *l2* second lateral nerve branch, *l3* third lateral nerve branch, *ps* posterior septum, *sg* segmental ganglion.

forms one segmental ganglion in each segment. The branching type of the lateral nerves in these segments is usual in Tubificidae, and each pair of the branches runs out laterally from about the middle of the ventral cord in the respective segment and innervates the integument of the body. Each segmental ganglion in these segments lies also near the posterior septum of the respective segment. In most segments posterior to Segment V, the ventral cord branches off three pairs of lateral nerves and forms one segmental ganglion in each segment. Each segmental ganglion lies in the posterior one-fourth of the ventral cord in each segment. The first two pairs of the lateral nerves are found anterior to the ganglion and the third pair posterior to the ganglion in each segment. These lateral branches, however, are not well-developed in the posterior segments.

VII. BLOOD-VASCULAR SYSTEM

The Dorsal Vessel. A large dorsal vessel begins at Segment V. At the portion anterior to Segment V, the dorsal vessel divides into paired branches. Each of these branches runs through the dorso-lateral side of the anterior alimentary tract, and reaches the prostomium. In Segments V-X, the dorsal vessel runs longitudinally a little to the right side of the median dorsal line of the alimentary tract (Fig. 16). At the anterior end of Segment XI, the dorsal vessel unites with the supra-intestinal vessel and then forms the median dorso-supra-intestinal vessel, first named by E. NOMURA ('26) in the case of *Tubifex hattai*. The so-called dorso-supra-intestinal vessel runs posteriorly along the median dorsal side of the intestine and is covered with the chloragogue coat. This dorsal vessel including dorso-supra-intestinal vessel is the thin-walled blood vessel without valve, measuring about 80μ in maximum diameter of the cross section, and gives off as branches one pair of lateral commissures in most segments.

The Ventral Vessel. The anterior ends of the ventral vessel, dividing also into two lateral branches, commence at the prostomium. Each of these branches runs posteriorly through the ventro-lateral side of the brain, of the buccal cavity and of the pharynx, and they unite together at the ventral side of the oesophagus in Segment V, a large ventral vessel being formed there. The ventral vessel runs further posteriorly along the median ventral side of the alimentary tract (Fig. 16). Besides giving off the capillary vessels as branches to septal sacs in Segments

VI and VII, the ventral vessel unites with pairs of the lateral commissures in most segments and with the intestinal hearts in Segments VIII and IX. This ventral vessel is a thin-walled blood vessel without valve, the diameter of this vessel being a little smaller than that of the dorsal vessel. According to the description of *Tubifex hattai* by E. NOMURA ('26), the atrophy of the intermediate ventral vessel is found in Segments VIII and IX. Such an atrophy, however, is not found in the ventral vessel of *Tubifex tubifex* reported by VEJDOVSKY ('84) and by DIXON ('15). The ventral vessel of the present species is similar to that of *Tubifex tubifex*.

The Lateral Commissures. The above-mentioned anterior branches of the dorsal and of the ventral vessel are nothing but the modified lateral commissures in the anterior portion of the body. The first pair of the typical lateral commissures, surrounding the lateral wall of the alimentary tract, runs out from the dorsal vessel of Segment V downwards in a convoluted pathway, and unites with the ventral vessel in the posterior level of Segment V. In an almost similar pathway, the second pair runs out from the dorsal vessel of Segment VI and unites with the ventral vessel at the beginning of Segment VII. The third pair, arising from the dorsal vessel of Segment VII, runs downwards along the lateral wall of the alimentary tract in an S-shaped pathway, and unites with the ventral vessel in that segment (Fig. 16). In Segments VIII and IX, no lateral commissure is present. The lateral commissures in Segments X-XII are of a different type developed specially: those in Segment X send out many branches to the wall of the sperm-sac, and those in Segments XI and XII to the wall of the ovisac respectively. The lateral commissures in Segments XIII-XV are the connexions between the dorsal and the ventral vessel, surrounding again the lateral wall of the intestine in each segment. The remaining pairs of the lateral commissures posterior to Segment XV form the so-called integumentary commissures in each segment, and they run downwards along the curvature of the body-wall from the dorsal to the ventral vessel. At the terminal portion of the body, these integumentary commissures are well-developed and form the integumentary network of the blood vessels.

The Intestinal Network of the Blood Vessels. The intestinal network of the blood vessels commences at the posterior half of Segment V. From this network, the supra-intestinal vessel appears at about the middle of Segment VI and the sub-intestinal vessel at about the middle of Segment VIII. In Segments V-XII, the capillary blood vessels constituting the intestinal network, invade among the interspaces of epithelial cells of

the intestinal wall. In Segments XIII–XX, these capillary vessels develop suddenly to large vessels, which come to form a conspicuous network around the intestinal wall. At the portion posterior to Segment XX, however, the intestinal network becomes gradually indistinct and disappears finally at about the level of Segment I.

The Supra-intestinal Vessel. The supra-intestinal vessel attaches closely to the dorsal side of the intestinal wall of Segments VI–X. In Segments VIII and IX, the supra-intestinal vessel gives off as branches a pair of the intestinal heart in each segment (Fig. 16). The supra-intestinal vessel is covered with the chloragogue coat, the diameter of this vessel being about equal to that of the dorsal vessel.

The Sub-intestinal Vessel. The sub-intestinal vessel runs posteriorly towards the level of Segment XXX, attaching closely to the median ventral wall of the intestine. The sub-intestinal vessel connects also with the ventral vessel by one pair of small bridges in each segment posterior to Segment X. These paired bridges between the sub-intestinal and ventral vessels may be called the ventro-sub-intestinal connectives.

The Intestinal Hearts. Two pairs of the intestinal hearts are found in this species. Each heart is a bulky blood-vessel, forming the connexion between the supra-intestinal vessel and the ventral vessel (Fig. 16). The first pair of hearts arises from the supra-intestinal vessel at about the middle of Segment VIII, and then encircles obliquely the lateral wall of the intestine in a downward direction to unite with the ventral vessel at about the middle of Segment IX, the maximum outline of this heart in

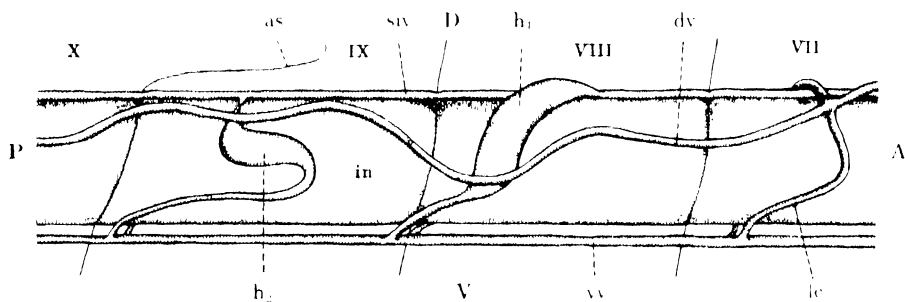


Fig. 16. Reconstructive scheme viewed from right side, illustrating intestinal hearts and their allied blood vessels. $\times 50$. A anterior side, D dorsal side, P posterior side, V ventral side of body, VII, VIII, IX and X No. of segment; as anterior sperm-sac, dv dorsal vessel, h_1 first heart, h_2 second heart, in intestine, lc lateral commissure of blood vessel in Segment VII, sv supra-intestinal vessel, vv ventral vessel.

the cross section being assumed approximately to be an elliptical shape and its two diameters being measured with $200 \times 100 \mu$ (Fig. 17 A). The second pair of hearts arises from the supra-intestinal vessel at about the middle of Segment IX and runs in an S-shaped pathway along the lateral wall of the intestine to unite with the ventral vessel at about the end of

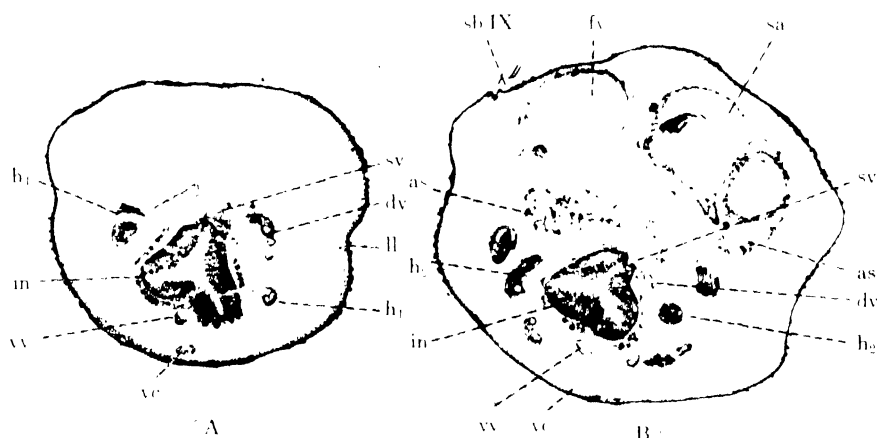


Fig. 17. Two photomicrographs, showing first A and second B pair of intestinal hearts in transverse sections through Segments VIII and IX $\times 50$ as anterior sperm-sac, dv dorsal vessel, fv floating vesicle, h_1 first heart, h_2 second heart, in intestine, ll lateral line, sa spermathecal ampulla, sb IX seta-bundle of Segment IX, sv supra-intestinal vessel, vc ventral nerve cord, vv ventral vessel.

Segment IX. The maximum diameter of the second heart in the cross section is a little smaller than that of the first heart (Fig. 17 B). The wall of these hearts with neither special thickening nor valve, consists of two layers, the outer cellular peritoneum and the inner intima.

VIII. GENITAL SYSTEM

The male genital organs of the present species are one pair of testes, of sperm-ducts, of penes and of prostate glands, in addition to the anterior and posterior sperm-sacs occupying the median position of the body (Fig. 2).

The Testes. The testes are paired solid masses of long ellipsoidal shape, lying along the body-wall on both the lateral sides of the intestine of Segment X. The anterior end of each testis attaches to the posterior face of Septum IX/X, and its posterior end extends to about the middle of Segment X. No special investment is found around the testis, and the male cells in early spermatogenetic stages are liberated from the

postero-dorsal side of testis into the cavity of sperm-sacs.

The Sperm-Sacs. The sperm-sacs are the large coelomic pouches expanding out from the median dor-al half of Segment X. The anterior sperm-sac is formed by the anterior diverticulum of Septum IX X and extends to the anterior end of Segment IX. The posterior sperm-sac is formed by the posterior diverticulum of Septum X XI and extends to the posterior end of Segment XIII. The wall of these sperm-sacs is equal to

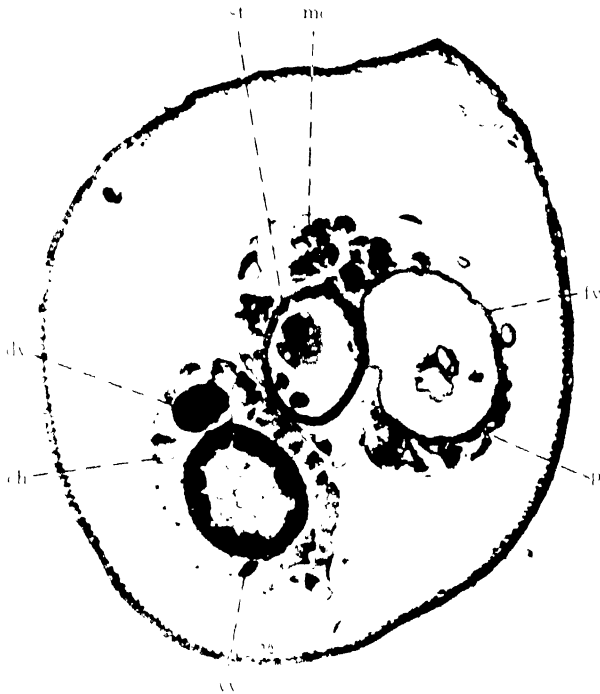


Fig. 18. Photomicrograph, showing posterior sperm-sac, spermathecal ampulla and floating vesicle in transverse section through beginning of Segment XII. $\times 170$. *ch* chloragogue cell, *dv* dorso-supra-intestinal vessel, *fe* floating vesicle, *me* male cell in sperm-sac, *sp* posterior sperm-sac, *st* spermathecal ampulla, *va* ventral vessel.

the other septa in structure, although the blood supply from the branches of the dorsal and ventral blood vessels in Segment X is observed in the wall of the posterior sperm-sac. The male cells liberated from the testes complete their spermatogenesis inside these sperm-sacs. In fact, many male cells in various spermatogenetic stages including the ripe spermatozoa are found there (Fig. 18). A dimorphism of the ripe sper-

matozoa was reported by DIXON ('15) in the case of *Tubifex tubifex*. On the contrary, GATENBY ('16) denied the dimorphism of ripe spermatozoa in the same species. The present writers are under impression that the so-called dimorphism is not found among the fully matured spermatozoa of *Tubifex (Peloscolex) nomurai*.

The Sperm-Ducts. The sperm ducts are the paired organs, lying on both the lateral sides of the intestine of Segment XI. Each sperm-duct consists of a sperm-duct funnel, a long vas deferens, and an atrium.

(a) THE SPERM-DUCT FUNNELS. Each sperm-duct funnel attaches to the respective ventro-lateral part of the anterior face of Septum X XI, and opens widely along the curvature of the body-wall. The inner face of the funnel is furnished with fine cilia, and the outer face is covered with connective tissue derived from the peritoneum. Inside this funnel, many ripe spermatozoa are arranged in definite order, with their head towards the entrance of the duct.

(b) THE VASA DEFERENTIA. The posterior end of each sperm-duct funnel is continuous to a slender, convoluted vas deferens. Each vas deferens lies generally along the dorso-lateral side of the intestine of Segment XI, and measures about 1.5 mm. in whole length. The posterior

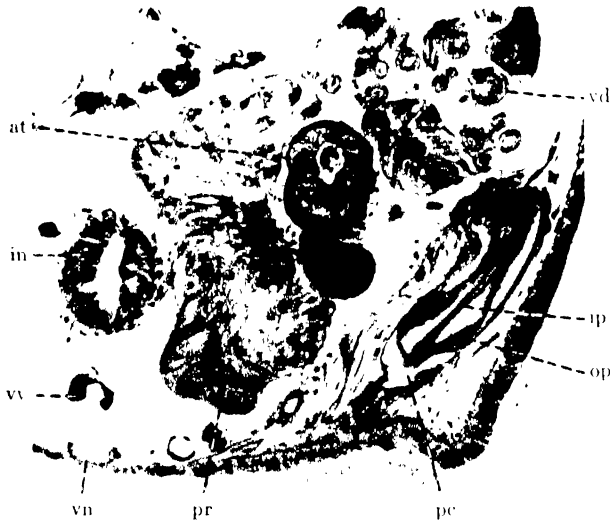


Fig. 19. Photomicrograph, showing vas deferens, atrium, penis and prostate gland in transverse section through middle portion of Segment XI. $\times 120$. *at* atrium, *in* intestine, *ip* inner wall of penis, *op* outer wall of penis, *pc* penial chamber, *pr* prostate gland, *vd* vas deferens, *vn* ventral nerve cord, *vv* ventral vessel.

end of each vas deferens reaches to the posterior two-thirds of Segment XI (Fig. 19). Each vas deferens frequently winds to the opposite side, owing to the extensive convolution. According to the descriptions given by DIXON ('15) and by NOMURA ('26), the inner face of vas deferens is constituted of the proximal ciliated and distal non-ciliated epithelia in the case of *Tubifex tubifex* and of *Tubifex hattai* respectively. In *Tubifex (Peloscolex) nomurai*, however, the present writers could observe the fine cilia through the whole inner face of vas deferens. The cytoplasm of the ciliated cells, constituting the epithelial wall of vas deferens, are often vacuolated, and their nuclei show peculiar sausage shapes. Furthermore, the thin cell-layer covers this ciliated wall of vas deferens.

(c) THE ATRIA. The posterior end of each vas deferens is continuous to an atrium, which consists of the anterior, ampullar portion and the posterior duct-portion. The ampullar portion of each atrium lies on the dorso-lateral side of the intestine, and measures about 0.5 mm. in length. This ampullar portion is thick-walled, measuring about 150μ in diameter and about 20μ in caliber (Fig. 19). Three layers are discriminated in this thickened wall. The innermost layer is composed of columnar cells without cilia. The granules, contained in some of these columnar cells, are densely stained with Delafield's haematoxylin. The middle layer is composed of thick circular muscles and is stained with eosin. The outermost layer is the covering of connective tissue cells. The prostate gland opens at about the middle of the dorsal wall of this ampullar portion. The duct-portion of each atrium measures about 0.2 mm. in length and $30\text{--}50\mu$ in diameter. The wall of this duct-portion is not so thick as that of the ampullar portion. The innermost layer of this duct-portion is composed of non-granulated cells.

The Prostate Glands. The prostate glands are paired large organs, occupying the posterior half of Segment XI. Each gland is solid and irregularly shaped, and opens to the ampullar portion of the atrium (Fig. 19). Each cell constituting this gland has a dense cytoplasm, rich in fine granules, and the spherical nucleus, containing a distinct nucleolus.

The Penes. The posterior end of each atrium is continuous to a fusiform penis. This penis is kept in the penial chamber, which is formed by the upward invagination of the body-wall at the posterior one-fourth of Segment XI on the ventral setal line (Fig. 19). Therefore, the inner face of the penial chamber is lined with the cuticular layer. The outer wall of the penis is continuous to the wall of the penial chamber and is the outpushing of columnar hypodermal layer, which is destitute of cuticle.

The inner wall of the penis is continuous to the innermost layer of the atrium and forms the narrow lumen of duct up to the terminal extremity. The interspace is found between these outer and inner layers. The retractor muscle is well-developed at the basal portion of the penis. In a retracted condition, the penis measures 280–300 μ in length.

The female genital organs of the present species are one pair of spermathecae, of ovaries and of oviducts, in addition to an ovisac occupying the median position of the body (Fig. 2).

The Spermathecae. Each spermatheca consists of an ampullar portion, a duct proper and a special gland.

(a) THE SPERMATHECAL AMPULLAE (Fig. 18). The ampulla of each spermatheca lies in the cavity of sperm-sacs, and extends from Segment X to XII. This ampulla is sausage-shaped and measures 1–1.2 mm. in length, a few diverticula being often added there. The wall of the ampulla is thick, measuring 8–10 μ , and consists of the inner and outer layers. The inner epithelium is composed of cubical cells and the outer of flattened cells. In the fully matured specimen, many spermatophores are present inside this ampulla.

(b) THE SPERMATHECAL DUCTS. The duct of each spermatheca arises at the anterior end of the ampullar portion, and opens externally at the middle of Segment X on the respective ventral setal line (Fig. 20 A). The duct measures 0.2–0.3 mm. in whole length, and diminishes gradually in diameter towards the external opening. The wall of the duct consists of three cell-layers. The innermost layer is composed of columnar or club-shaped cells and is non-ciliated. The middle is a circular muscle layer and the outermost is a peritoneal layer.

(c) THE SPECIAL GLANDS (Fig. 20). A special gland is observed at the terminal portion of each spermathecal duct of the present species. The gland apparatus in connexion with genital setae has been found in some species of Gen. *Tubifex* and of Gen. *Ilyodrilus*, viz., *Tubifex barbatus* (GRUBE), *Tubifex illustris* DITLEVSEN, *Tubifex insignis* (EISEN), *Tubifex ochridanus* HRABE, *Ilyodrilus sodalis* EISEN, etc. The spermathecal gland apparatus of the present species, however, differs considerably in appearance from that of the other species. It consists of three portions, viz., a glandular cell-mass, its receptor and a muscle ring between the two. The nucleus of each gland cell occupies the distal position entirely. The receptor opens to the lumen of the terminal portion of the spermathecal duct through a slit-like opening. The wall of the receptor consists of

two layers, the inner thin epithelium and the outer well-developed muscle layer. The muscle ring between the glandular cell-mass and its receptor is presumed to control the secretion from the gland.

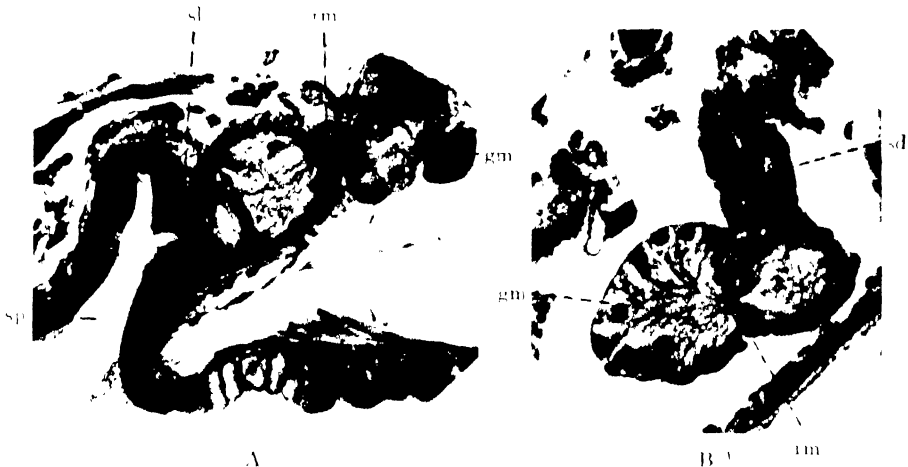


Fig. 20. Two photomicrographs, showing terminal portion of spermathecal duct and spermathecal gland apparatus in longitudinal A and transverse B sections $\times 250$. *gm* glandular cell-mass of spermathecal gland, *rm* muscle ring of spermathecal gland, *sd* spermathecal duct, *sl* opening of spermathecal gland, *sp* pore of spermathecal duct.

The Ovaries. The ovaries are paired solid masses of somewhat larger ellipsoidal shape than the testes, and lie along the body-wall on both lateral sides of the intestine of Segment XI. The anterior end of each ovary attaches to the posterior face of Septum X/XI, and its posterior end extends to about the middle of Segment XI. Dixon ('15) reports a central, non-nucleated, cytoplasmic core surrounded by developing oöcytes in the mature ovary of *Tubifex tubifex*. In *Tubifex (Peloscolex) nomurai*, such a cytoplasmic core in the centre of the ovary was not observed, but the dorsal and posterior elements of a given ovary developed earlier than the others.

The Ovisac. The ovisac is a large dorso-median pouch, which is formed by the posterior diverticulum of Septum XI/XII and is telescoped the posterior sperm-sac. The posterior end of the ovisac reaches to the middle of Segment XIV. This ovisac is filled up with many developing ova containing yolk granules.

The Oviducts. The oviducts are paired organs, lying at both ventral parts of the intersegment between Segments XI and XII. Each oviduct

consists of a oviduct funnel and a short duct proper.

(a) **THE OVIDUCT FUNNELS.** Each oviduct funnel opens at the anterior face of Septum XI/XII near the ventral body-wall, and consists of two layers. The inner layer is constituted of cubical cells furnished densely with fine cilia. The outer face of this ciliated epithelium is covered with a peritoneal layer.

(b) **THE DUCT PROPER.** The posterior end of each oviduct funnel is continuous to the duct proper. This duct pierces the Septum XI/XII and runs ventrally along the posterior face of that septum. In the fully matured specimen, the terminal end of this duct opens externally at the intersegmental furrow between Segments XI and XII on each ventral setal line.

Besides the male and female organs mentioned above, several thin-walled vesicles lie freely inside the anterior and posterior sperm-sacs. The function of these vesicles is unknown and are tentatively named "floating vesicles" by the present writers.

The Floating Vesicles. Usually 7-9 floating vesicles are found in the sperm-sacs of mature specimen, and a less number of them in those of immature specimen. Each vesicle is filled by a fluid substance taking a light purple stain with Delafield's haematoxylin. Each vesicle is of a sausage-shape, about similar to that of the spermathecal ampulla. The wall of each vesicle is constituted of flattened cells (Fig. 18). The posterior end is prolonged into a slender, convoluted duct. The end of this duct terminates freely inside the sperm-sacs and has no connexion to any other organs nor with the external opening. The lumen of this duct is filled with yellowish substance, supposed to be a metamorphosed fat-body. In the case of *Haplotaxis gordioides* (HARTM.), LEYDIG (1865) reports hollow and complicated vesicles containing a metamorphosed fat-body. Moreover, he states that these vesicles may be some modifications of the spermatheca which are filled up with sperms first by the copulation. No such vesicular organ has been found in any other species of aquatic oligochaetes. This floating vesicle of *Tubifex (Peloscolex) nomurai* seems to be somewhat similar to LEYDIG's vesicle of *Haplotaxis gordioides*. The writers are of opinion, however, that the floating vesicles of the present species, having a wall of unicellular structure, are neither a modification of spermathecal ampulla nor an artifact producing inside the sperm-sacs.

GENERAL CONSIDERATIONS AND CONCLUSION

Judging from the preceding detailed description, the present species should undoubtedly be classified into the *Tubifex*-group of the Family Tubificidae. More than ten species of this group have been so far obtained from deep lakes by MICHAELSEN and by HRABĚ. The present species, however, is distinguished clearly from these known species by the following characteristics:

1) *The floating vesicles.* Such vesicles have not been found in any other species of the Family Tubificidae.

2) *The special gland of the spermathecal ducts.* In some species of the *Tubifex*-group, the gland apparatus has been found at the terminal portion of each spermathecal duct. Moreover, in these instances, the genital setae have also been present invariably together with the gland apparatus of the spermathecal duct. The present species, however, has no genital setae notwithstanding the presence of the gland apparatus of the spermathecal duct.

3) *The two pairs of the intestinal hearts.* BEDDARD (1895) and STEPHENSON (1930) report that the presence of two pairs of intestinal hearts is a common characteristic of the Genera *Limnodrilus* and *Clitellio* in the Family Tubificidae. On the other hand, various other types of heart have been reported among the members of the *Tubifex*-group, two pairs of intestinal hearts among them being observed for the first time in the present species. The writers are of opinion that this type of heart is one of the important specific characteristics of the present species of the *Tubifex*-group.

According to STEPHENSON's system (1930), the *Tubifex*-group was classified into three Genera, viz., *Tubifex*, *Peloscolex* and *Ilyodrilus*. In *Tubifex* and *Peloscolex*, the length of vas deferens is longer than or equal to that of atrium, while, in *Ilyodrilus*, the length of vas deferens is shorter than that of atrium. Furthermore, in *Peloscolex*, the papillae are present, while, in *Tubifex*, the papillae are not present.

In the present species, each vas deferens is comparatively long, being about two or more times as long as each atrium. Small sized papillae are also present, and those only in the clitellum, being densest in its ventral half. These poorly-developed papillae in the present species are very remarkable. The present species may be classified into a new species of the Genus *Peloscolex* after STEPHENSON's system. In already known species of *Peloscolex*, however, various kinds of well-developed papillae

have been reported so far. MICHAELSEN ('33) figured the cellular structure of the "sensory papillae" in the case of *Peloscolex wereschtschagini*. RANDOLPH ('92) reports, in the sensory papillae of *Peloscolex plicatus* and *Peloscolex veltinus*, that several fine hairs were present. STEPHENSON ('22) reports, in the secretory papillae of *Peloscolex benedeni*, that small granules were present. And also, these well-developed papillae are found in most segments of both mature and immature specimens, and are always arranged in a few rings. For exception, poorly-developed papillae are only found in *Peloscolex insularis*. As stated already, the papillae of the present species which have been observed only in mature specimens, are certainly simpler in structure and fewer in number in comparison with those of the known species of *Peloscolex*. Thus, the writers are of opinion that the distinction of *Peloscolex* from *Tubifex* after STEPHENSON's system is unreliable and difficult, and that the Genus *Tubifex* including the Subgenus *Peloscolex* is appropriately opposed to the Genus *Ilyodrilus* among the members of *Tubifex*-group, after MICHAELSEN's (1900) and POINTNER's (1911) systems.

In conclusion, the present species appears to be a new intermediate form between the Genus *Tubifex* and the Genus *Peloscolex*, after STEPHENSON. Therefore, the writers prefer to follow MICHAELSEN's system and beg to introduce as a new species, *Tubifex (Peloscolex) nomurai*.

PUBLICATIONS REFERRED TO

- BEDDARD, F. E., 1895. A Monograph of the Order Oligochaeta. Oxford.
- BENHAM, W. B., 1903 a. A Note on the Oligochaeta of the New-Zealand Lakes. Trans. Proc. N. Z. Inst., 36.
- BENHAM, W. B., 1903 b. On Some New Species of Aquatic Oligochaeta from New-Zealand. Proc. Zool. Soc. London, 2.
- CERNOSVITOV, L., 1926. Eine neue Variation des *Tubifex tubifex* MÜLL. Zool. Anz., 65.
- DITLEVSEN, A., 1904. Studien an Oligochaeta. Z. wiss. Zool., 77.
- DIXON, G. C., 1915. *Tubifex*. L. M. B. C. Memoir 23, London.
- HRABĚ, S., 1931. Die Oligochaeten aus den Ochrida und Prespa. Zool. Jahrb., Abt. Syst., 61.
- LEYDIG FR., 1865. Über *Phreoryctes menkeanus* HOFM. nebst Bemerkungen über den Bau anderer Anneliden. Arch. Mikrosk. Anat., 1.
- MICHAELSEN, W. 1900. Das Tierreich. Eine Zusammenstellung und Kennzeichnung der rezenten Tierformen. 10. Lief., Vermes, Oligochaeta, Berlin.
- MICHAELSEN, W. 1905. Wissenschaftliche Ergebnisse einer Zoologischen Expedition nach dem Baikalsee unter Leitung des Professor ALEXIS KOROTNEFF in den Jahren 1900-1902. Die Oligochaeta des Baikalsees. Berlin.
- MICHAELSEN, W. 1933. Eine Panzeroligochaet aus dem Baikalsee. Zool. Anz., 102.

- MICHAELSEN, W. & L. JOHANSSON, 1909. Die Süßwasserfauna Deutschlands., Heft 13, Oligochaeta und Hirudinea. Jena.
- NAGASE, I. & E. NOMURA, 1937. On the Japanese Aquatic Oligochaeta, *Criodrilus miya shitai*, n. sp. Sci. Rep. Tôhoku Imp. Univ., Biol., 11.
- NASSE, D., 1882. Beiträge zur Anatomie der Tubificiden. Bonn.
- NOMURA, E., 1913. On two Species of Aquatic Oligochaeta, *Limnodrilus gotoi* HATAI and *Limnodrilus willeyi* n. sp. Jour. Coll. Sci. Imp. Univ. Tokyo, 35.
- NOMURA, E., 1926. On the Aquatic Oligochaete, *Tubifex hattai* n. sp. Sci. Rep. Tôhoku Imp. Univ., Biol., 1.
- POINTNER, H., 1911. Beiträge zur Kenntnis der Oligochaetenfauna der Gewässer von Graz. Z. wiss. Zool., 98.
- RANDOLPH, H., 1892. Beiträge zur Kenntnis der Tubificiden. Jen. Zeits. Naturw., 27.
- SMITH, F., 1900. Notes on Species of North American Oligochaeta III. List of Species found in Illinois, and Descriptions of Illinois Tubificidae. Bull. Ill. St. Lab. Nat. Hist., 5.
- STEPHENSON, J., 1930. The Oligochaeta. Oxford.
- STOLTE, H. A., 1933, '35, '38. Bronns Klassen und Ordnung des Tierreichs. 4 Bd. Vermes, 3 Abt., Annelides, Oligochaeta., Leipzig.
- VEJDovsky, F., 1884. System und Morphologie der Oligochaeten. Prag.

THE LIFE HISTORY OF A NEW CYNIPID FLY, *KLEIDOTOMA JAPONICA*, N. SP.

By

KAORU HUZIMATU

Biological Institute, Tôhoku Imperial University, Sendai

(With Plates XIX XXIII and eleven text-figures.)

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I. INTRODUCTION

Most members of the Family Cynipidae are known as gall-makers, and their biology and bionomics have been reported upon by many writers. But in the same family there are also entomophagous groups expressed by such subfamilies as Eucilinae, Figitinae, and Charipinae. The Eucilinae and the Figitinae are parasitic chiefly upon Diptera, and the Charipinae are known to attack aphids and coccids. During the life history of the entomophagous Cynipidae, hypermetamorphosis is known to occur as in some other parasitic Hymenoptera, but our knowledge regarding this subject seems to be rather scanty. KEILIN and PLUVINEL (1913), in their studies of *Eucoila*, were the first to describe the primary larval form of the entomophagous Cynipidae. HAVILAND (1923) gave some account on the early larval stage of *Charips*, and JAMES (1927) made a report on the same of *Cothonaspis*, *Kleidotoma* and *Figites*, and contributed many new and interesting data. But no detailed reports on the metamorphosis and the biology of the entomophagous Cynipidae have yet been published and many problems which should be studied have been left untouched. In this paper I shall deal with a new species of Japanese *Kleidotoma* making special reference to its larval forms in their early stage and also to its life history. The new species which I am going to describe in this paper is named *Kleidotoma japonica* and is parasitic upon the larva and the pupa of a dipterous fly, *Scatella calida* which lives in hot springs.

Before proceeding further, I wish to express my hearty thanks to Prof. Dr. SANJI HÔZAWA under whose direction and encouragement my investigations have been made. I am also greatly indebted to Prof. Dr. T. ISHII and Prof. Dr. Y. OKADA for their helpful suggestions and for their kindness to me in the course of the identification of the present

species. I wish also to take this opportunity to thank Mr. S. KINOSHITA, and Mr. K. TUTUI for their assistance with regard to the literature concerning this species. I am also grateful to Dr. R. K. START, Rev. P. S. C. POWLES, Assist. Prof. Dr. I. MOTOMURA and Mr. M. KATO for all their kindness expressed to me during the course of the present study.

II. MATERIALS AND METHODS

The observations were made during the period extending from the beginning of June to the end of July, 1937, at Tubame hot spring, Niigata Prefecture. Tubame hot spring is located at the foot of Mt. Myôkô. *Kleidotoma japonica* is parasitic upon the larva and the pupa of *Scatella calida*, a fly which lives in hot springs of acidic nature. The larvae and pupae of this fly infested by *Kleidotoma japonica* were collected in shallow hot spring water, and the adults of *Kleidotoma* were chiefly reared in the laboratory, though some were obtained in the field. Dissections were made under a microscope in order to obtain the eggs and the larvae of the parasite from the body-cavity of the host. For the morphological observation of the eggs and larvae living materials were chiefly used. The histology of the gut was studied by cutting sections. Carnoy's and Boin's fluids were used as fixatives. The sections were stained by Delafield's haematoxylin and by Heidenhain's haematoxylin, being counterstained respectively with eosin and orange G. In measuring the eggs, the larvae and the pupae, the living specimens were used.

III. NOTES ON THE HOST FLY, *SCATELLA CALIDA* MATSUMURA

The larvae of *Scatella calida*, live in shallow streams and pools of hot springs of acidic nature (Text-fig. 1, 2). The adult flies swarm on the surface of the water of the hot springs, and also on the surfaces of stones and pieces of wood half submerged in the water of the same. The adult is a small fly, measuring about 4 mm. in the total length. The egg is an elongated oval in shape, measuring 1.1 mm. in length and 0.3 mm. in breadth. The egg is milky-white in colour, and its surface shows a minute network of uniform appearance. The eggs are laid usually in a mass containing from 20 to 30, being rarely deposited one by one. The egg masses thus laid are usually attached to such objects as stones, pieces of wood and dumps of moss half submerged in the hot spring. The larva, newly emerged from the egg, is a small maggot, measuring about 1.5 mm. in length. There are three stages in the larval



TEXT-fig. 1. A very typical habitat of *Scatella calid* and *Kleidotoma japonica*. A small hot spring flowing from a rock fissure can be seen.



Text-fig. 2. A hot spring pool where *Scatella calid*, *Kleidotoma japonica* and *Stratiomyia japonica* are found.

history, as shown distinctly by the measurements of the cephalopharyngeal skeletons. The full-grown larva measures about 5 mm. in length. It is a maggot of a milky-white colour and of cylindrical form with the dorsal and the ventral surface slightly flat. The larvae feed chiefly upon diatoms growing in the hot springs. The puparium measures about 4.5 mm. in length. The colour of the puparium is a yellowish-brown at first, but it turns gradually to a dark brown. The larvae and the adults may be seen in great numbers in June, July and August, but as the autumn advances, they gradually decrease in number. Most of the flies pass the winter in the forms of larva and pupa, though some attain to the adult form. The temperature of the hot springs in which the larva of this fly live, ranges from 30°C to 42°C and the pH of the springs, ranges from 4.0 to 6.5.

Scatella calida has also been found in the hot springs at Jōzankei (MATSUMURA, 1917), and at Sarukura (OKADA, 1936).

The female of *Kleidotoma japonica* deposits one egg into the body-cavity of the larva of *Scatella calida*, when that larva is in the first or the second stage. The host-larva thus infested continues its development without showing any obstruction caused either by the egg laid in it, or by the young larva when hatched from the egg, and it becomes full-grown and then pupates. When the full-grown larva of the host has pupated, the parasite devours the host pupal-body completely, leaving only the puparium, and then it pupates itself in the puparium of the host. It is difficult to distinguish by external appearance whether the larva of *Scatella calida* is infested or not by *Kleidotoma*. But when the host-larvae are examined under a microscope, the egg or the early-stage larva contained in the body-cavity may be detected under the skin of the host.

IV. DESCRIPTION OF *KLEIDOTOMA JAPONICA*, N. SP.

PLS. XIX-XX, Figs. 1-10

Kleidotoma japonica, n. sp.

Female. Shining black in general. Antennae dark brown. Wings hyaline, veins dark brown. Legs brown except coxa and femur which are dark brown.

Head slightly longer than wide; cheek with a fine furrow and soft hairs; compound eyes, nearly round; ocelli arranged in a slightly flattened triangle. Mandibles broad, with bristles on the surface; tridentate, the apical tooth the largest. Maxillae with large fleshy galea; stipes elongated

oval, with hairs on the surface; maxillary palpi composed of five joints; the basal joint the smallest; the second and the third joints short and thick; the fourth and the fifth joints nearly equal in length, and about twice as long as the second or the third joints; the fifth joint with bristles and a small papilla, one of those bristles very long. Labial palpi composed of two joints, of nearly equal length; the second joint with short bristles and papillae at its apical end; glossa and paraglossae with hairs. Antennae 1.16 mm. in length, about two-thirds as long as the body-length, each joint with short hairs and some slight ridges. Antennae composed of thirteen joints, measurement of each joint is shown as follows.

No. of joints.	1	2	3	4	5	6	7	8	9	10	11	12	13
Length of joints in mm.	0.130	0.060	0.120	0.065	0.065	0.065	0.075	0.080	0.080	0.095	0.095	0.095	0.120

The first joint large, slightly curved, with a narrow base and truncated broad end; the second globular, nearly as long as broad; the third long and slender, twice as long as the second; the fourth much shorter, nearly half as long as the third; the seventh half-clubbed; the eighth to thirteenth joints distinctly clubbed; the thirteenth larger and broader than the previous joints.

Thorax 0.65 mm. in length and 0.40 mm. in breadth. Mesonotum convex above, with a few greyish hairs on the surface; parapsidal furrows obsolete. Scutellum longitudinally striated, with six to seven bristles on each margin. The cup longish oval, pointed at the base and rounded at the apex; with a small round depression near the posterior margin and a pair of long bristles on the middle part; basal fovea large, deep, separated by a sharp keel which runs from the cup.

Abdomen 0.77 mm. in length and 1.36 mm. in breadth, compressed laterally with no hairs; the second segment large and long, occupying about three-fourths of the total length of the abdomen; the base of the abdomen with thick tufts of yellowish brown hair on each side.

Fore wing 1.60 mm. in length, slightly longer than the abdomen, the margin thickly fringed with long hairs, the surface covered densely with short hairs; the apex almost truncated, but very slightly incised; radial cellule elongated, in length nearly two and a half times the breadth; radial cellule opened at the fore margin, the under margin of the cellule highly angled; the second abscissa of radius about half as long again as the first; cubitus incomplete, appearing a light brown line; median vein obsolete. Hind wing fringed with long hairs on the apex and on the

hind margin; the surface of the wing densely ciliated.

Fore legs slender, with greyish hairs; coxa truncated at the base, in length about twice as long as coxa at the middle part; tibia as long as femur, with a forked hook at the extreme end; the first joint of tarsi as long as the sum of the following three joints, with about twenty combs on the inner side; the fifth longer than the previous joint, with a pair of claws at the extreme end. Mid legs slightly longer than the fore legs; coxa elongated oval, in length about twice as long as the breadth, tibia with a pair of spurs at the apex. Hind legs much longer than the fore and the mid legs.

Length of body. 1.7 mm.

Male. — Antennae 1.70 mm. in length, slightly longer than the body-length; the number of joints fifteen; the third joint larger and thicker, about a half as long again as the fourth; the following joints not clubbed as those of the female.

Length of body. 1.60 mm.

Host. — The larva and the pupa of *Scatella calida* MATSUMURA.

Habitat. — Tubame hot spring, Niigata Prefecture.

Type in the author's collections.

V. BIOLOGICAL NOTES ON THE ADULT OF *KLEIDOTOMA JAPONICA*

The adults of *Kleidotoma japonica* are usually found near the shallow hot springs where the larvae of *Scatella calida* live. They are found most abundantly in places where moss-patches cover the rock, and little streams of water from the hot spring pass among them. The powers of flight of the adult fly are rather poor and the present writer hardly observed any of them flying about in the field. This would be due partially to the conditions of the environment which is very moist on account of the hot springs.

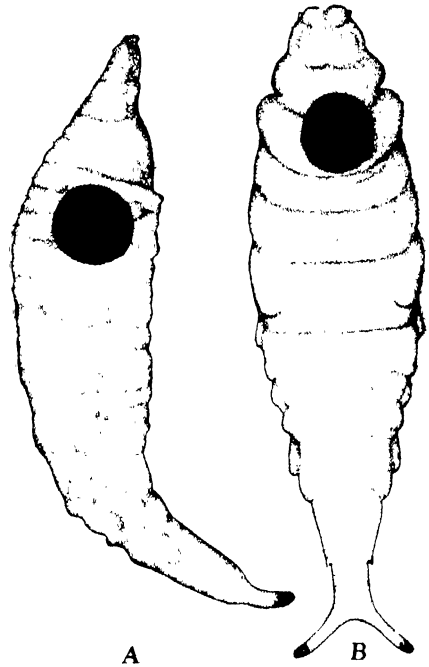
As will be seen from the description given above, the sexes of the adult fly may be easily distinguished by the lengths of the antennae. The mating takes place normally shortly after the emergence. The male chases the female, mounts on her body, and then bends the abdomen, inserting its tips in the genital opening of the female. The mating was observed in the field as well as in the glass tube within which the adults were reared from pupa. Copulation usually lasts for a few seconds. The writer was not able to observe the oviposition habit of the female, but it seems to be certain that she attacks the larva of *Scatella calida*

either in the first stage or in the second stage, and deposits one egg in it. The newly-laid egg is usually found in the body-cavity of the first or the second stage larva of the host. The adult of *Kleidotoma japonica* emerges from the puparium of the host-insect in which its pupal stage has been completed. When the adult fly emerges from the puparium, it makes for exit a circular hole in the wall of the puparium by means of its sharp mandibles. The hole for exit is generally circular in form, measuring about 0.7 mm. in diameter (Text-fig. 3, A, B). The hole is usually localized on the dorsal side near the anterior end of the puparium, but occasionally it is made on the lateral side.

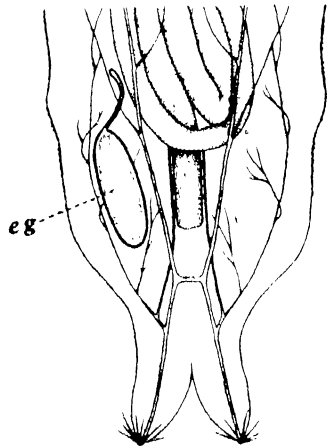
VI. THE EGG

(Pl. XXI, Figs. 14, 15)

The egg (Pl. XXI, Fig. 15) is an elongated oval in shape and is provided with a peduncle. It is 0.42 mm. long and 0.12 mm. broad. The peduncle is very narrow and is about half the length of the egg. The egg has a smooth surface and is milky-white in colour. The egg is laid in the posterior part of the body-cavity of the host, without being fastened to anything (Text-fig. 4). The eggs of the entomophagous Cynipidae which have been hitherto described are all pedunculated as are those of



Text-fig. 3. Puparia of *Scatella calida* showing holes of exit through which adults of *Kleidotoma japonica* have escaped. $\times 15$. A, lateral view. B, dorsal view.



Text-fig. 1. Posterior part of the larva of *Scatella calida*, showing the position of egg of *Kleidotoma japonica* deposited in the body-cavity. $\times 50$. eg egg.

the other Cynipidae and of certain Chalcids. The ovarian egg of *Eucoila keilini* is described by KEILIN and PLUVINEL (1913). It is an elongated oval in shape with a long peduncle measuring about twice the length of the egg-body. JAMES (1927) observed the egg of *Figites anthomyiarum* which had been laid in the body-cavity of a maggot. This egg was of a typical cynipid type with an elongated body and a peduncle as long as the body, but there was a constriction about the middle of the egg-body. In our species, the constriction of the egg-body was not observed. HAVILAND (1923) describes the egg of *Charips*, which is an oval body with a short peduncle. As the development of the embryo proceeds inside the egg, the peduncle of the egg gradually degenerates and becomes more or less withered. Just before the time of hatching, the young larva can be observed through the chorion of the egg. The head of the young larva is directed towards the anterior pole of the egg, where the peduncle is attached, three pairs of the thoracic appendage are arranged on the lateral side of the body, and the long caudal appendage is situated ventrally pointing towards the head (Pl. XXI, Fig. 1-1).

VII. THE LARVA

1. The First Stage

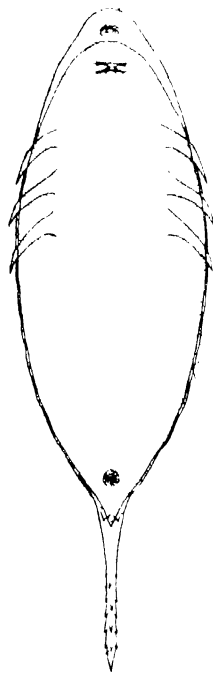
(Pl. XXI, Figs. 16-19)

As in other entomophagous Cynipidae hitherto known, hypermetamorphosis takes place in the life history of the present species. The larva of the first stage (Pl. XXI, Fig. 16) is a eucoiliform larva. This type of larva was first described by KEILIN and PLUVINEL in the case of *Eucoila keilini*, an endoparasite of *Pegomyia winthemi* MEIGEN. The distinguishing features of this type are the presence of three pairs of thoracic appendages and of a long caudal appendage. The larva of the first stage measures 0.55-0.65 mm. in length and 0.15-0.25 mm. in breadth at the broadest part. The body is milky-white in colour except the middle part which is yellowish on account of the gut content.

The larva possesses a head and a body with twelve segments, the posterior segments of which measuring 0.17 mm. at the level of the mouth parts. The oral opening lies on the surface of a large rounded proboscis and is surrounded by several papillae (Pl. XXI, Figs. 18, 19). The oral opening is nearly oval in shape, measuring 0.028 mm. in breadth.

Inside the proboscis there may be found a pair of long slender mandibles 0.017 mm. long. The mandible is strongly chitinized, and its

extreme end is sharply pointed. The mandibles are attached by large muscles with which they move freely. Behind the mandibles and in the middle of the oral cavity there may be seen a small beak-like sclerite. Its extreme end is sharply pointed and is curved ventrally like the beak of a bird. The beak-like sclerite is also attached by a large muscle by which it moves upwards and downwards. The mandibles and the beak-like sclerite may be the main organs by means of which the larva sucks the body-juice from the host. The head bears two pairs of sensory processes on the ventral surfaces, situated, one anteriorly, and the other posteriorly, to the mouth. The latter is furnished at the extremity with a transparent spot which seems to be sensory in function. Each of the first three body segments forming the thorax, has a pair of long appendages. The thoracic appendages measure 0.07-0.08 mm. in length and are furnished distally with minute setae. The body-segments diminish in breadth posteriorly, and the last segment bears a long caudal appendage. The caudal appendage measures 0.17 mm. in length and is armoured with a pair of prominent projections in the base and distally with a number of small setae. JAMES (1927) states that the thoracic appendages and the caudal appendage of eucoiliform larvae are of great use when the larva escapes from the egg. But the present writer did not have the opportunity of witnessing the hatching of the larvae from the egg and thus was not able to ascertain the function of the appendages. The anus opens on the postero-dorsal surface of the body near the base of the caudal appendage. The anus (Pl. XXI, Fig. 17) is large measuring 0.038-0.045 mm. diameter and is of peculiar structure being rounded by a ring of chitinous spines. A number of long spines concentrate from the periphery towards the centre of the anus and thus have an appearance somewhat like a spiracle. The larva is curved ventrally with the tail slightly bent to form a small angle with the abdomen. Just before the first moulting, the larva of the second stage can be seen through the old skin of the larva the first stage (Text-fig. 5).



Text-fig. 5. First stage larva just before the first moulting. Second stage larva can be seen through the skin of first stage larva.

2. The Second Stage

(Pl. XXII, Figs. 20-22)

The larva of the second stage (Pl. XXII, Fig. 20) is somewhat cylindrical in general form and measures 0.70-1.50 mm. in length and 0.18-0.40 mm. in breadth. The characteristic feature of this stage is the absence of the three pairs of thoracic appendages and of the long caudal appendage, both of which have been seen in the larva of the first stage or the eucoiliform larva. The head is rounded and the mouth opens on the ventral surface of the head (Pl. XXII, Fig. 21). The mouth-parts are quite different from those of the larva of the preceding stage. Namely, the rounded proboscis and the beak-like sclerite observed in the first stage cannot be seen in this stage. The mandible is longer and more thickly built, with the extreme end slightly pointed. Under the mandibles there are maxillae and labium, both of which are fused together. Using the mandibles, the maxillae and the labium, the larva takes the body-juice of the host into its pharynx. The body is jointed with twelve segments diminishing gradually in diameter posteriorly. The last segment is a little longer than the preceding one and bears a short caudal appendage (Pl. XXII, Fig. 22). The posterior end of the body is usually curved more or less dorsally and near the posterior end of the body there opens the anus dorsally. The anus of this stage is different from that of the first larval stage. It is transversely oval in shape, and it is not furnished with spines as in the case of the first larval stage, and it measures 0.04 mm. in length and 0.07 mm. in breadth. The larva of this stage can be found in the body-cavity of the third stage larva or in the pupa of the host insect.

3. The Third Stage

(Pl. XXII, Figs. 23, 24)

The larva of the third stage (Pl. XXII, Fig. 23) is nearly similar in general appearance to the larva of the second stage except as regards its size which is larger. It measures 1.70-2.30 mm. in length and 0.80-0.90 mm. in breadth. The body is an elongated oval in shape and the posterior end is less pointed than in the case of the second stage. The mouth-parts are nearly the same in structure as those of the second stage, but a pair of slight swellings of area antennalis are observable on the dorsal surface of the head (Pl. XXII, Fig. 24). The anus which opens

near the posterior end of the body is smaller than that of the preceding stage and is slit-like in shape. The larva of this stage was observed inside the pupal body of the host.

4. The Fourth Stage

(Pl. XXIII, Figs. 25, 26)

The larva of the fourth stage (Pl. XXIII, Fig. 25) is like a grub in form, measuring 1.9–2.8 mm. in length and 0.9–1.2 mm. in breadth. The mandibles are strongly chitinated, the apex being sharply pointed and dark brown in colour (Pl. XXIII, Fig. 26). The tracheal system is provided with three pairs of spiracles opening on the second, third and fourth segments. The anus appears as a slit-like opening as in the preceding stage. The larva of this stage can be seen in the puparium of the host insect.

5. The Fifth Stage

(Pl. XXIII, Figs. 27–30)

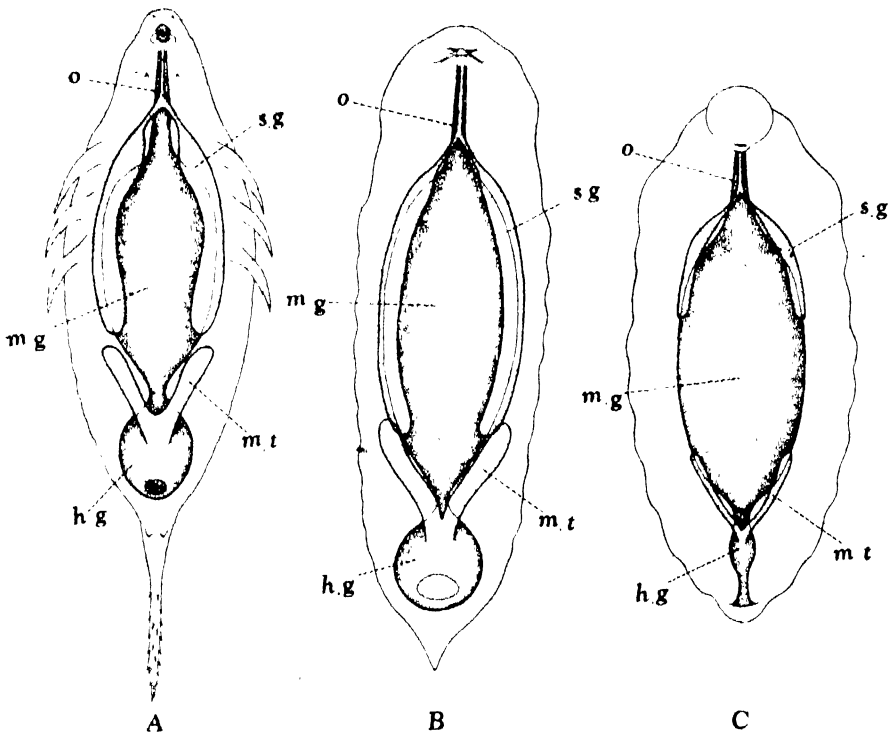
The larva of the fifth stage (Pl. XXIII, Figs. 27, 30) is a somewhat plump legless, hymenopterous grub of the usual type. The full-grown larva measures about 2.5 mm. in length and 1.0 mm. in breadth and the colour is milky-white with the integument smooth. It possesses a distinct head and twelve body segments. The head is nearly oval in shape, being about 0.30 mm. long and 0.35 mm. broad. From the front the head capsule is seen to be divided dorsally into two lobes. On the convex surface of each of these lobes is a small rounded elevation of area antennis representing the distal end of the antennal rudiment. The mouth opens on the ventral side of the head. The labrum is nearly oblong in shape, measuring 0.14 mm. in breadth and is furnished with a few papillae near the margin. The distal end of the labrum is free from the head capsule and its margin is slightly indented in the middle. The mandible is large and strongly chitinated, the colour being dark brown. It measures 0.07 mm. in length and bears from nine to ten denticles. The base of the maxilla is rounded and bears a small disk, upon which there are three minute papillae. The apex of the maxilla tapers and is directed inwards the mouth-opening. The labium measures about 0.06 mm. in breadth and bears a papilla on each side (Pl. XXIII, Fig. 30). Nine pairs of spiracles are present, one pair belonging to each segment from the second to the

tenth segment inclusive. They are located at the same level on each segment, about half-way between the dorsal mid-line and the ventral suture, and lie close to the anterior edges of their respective segments. The last abdominal segment bears the anus which appears as a slit-like opening. The full-grown larva having devoured completely the inside of the pupal body of the host, becomes motionless and experiences a short resting stage. This may be a so-called prepupal stage. In this stage the body of the larva becomes more elongated than that of the active full-grown larva. In this stage the second segment of the body is wide, provided with a voluminous protuberance on each side, the third and the fourth are rather narrow, while the fifth and the sixth increase in breadth, and the following segments diminish in breadth posteriorly. The prepupa casts its old skin and then pupates within the puparium of the host.

6. The Alimentary System

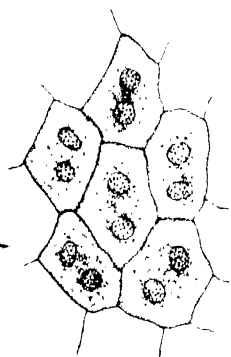
The mouth of the larva of the first stage is of suctorial type. The larva sucks the body-fluid of the host by means of its mandibles and beak-like organ. The alimentary canal (Text-fig. 6, A) comprises a short and slender fore-gut, a voluminous mid-gut and a swollen hind-gut. The oesophagus is a slender cylindrical tube which opens with a small valve to the mid-gut. The mid-gut is a voluminous sack occupying the greater part of the body-cavity. The hind-gut is globular in form, bearing a large lumen inside. This is enclosed by modified cells and opens exteriorly through the spined anus. As in the cases of other hymenopterous larvae of early stages, there is no communication between the mid-gut and the hind-gut. When the living larva of this stage is stained with neutral-red or methylene-blue, the hind-gut will be soon coloured with the stain readily entering through the anus before any other part of the body is affected. The salivary gland comprises a pair of slender cylindrical tubules extending from the second segment halfway down the body. At the second segment where the gland begins each gland opens cephalad into a thin-walled duct running to the head region, where it meets its mate from the opposite side. The two ducts above mentioned unite to form a common duct which opens to the mouth. The Malpighian tubules are two in number and are exceedingly short. They lie on each side of the hind-gut and communicates with it through a common opening.

The alimentary system of the larva of the second stage (Text-fig. 6, B) is of the same features and position as that of the first stage. But the

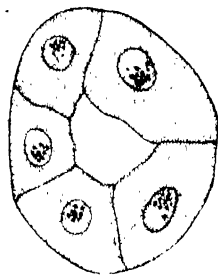


Text-fig. 6. Alimentary system of the larva. A. First stage larva. B. Second stage larva. C. Fifth stage larva. h.g. hind-gut; m.g. mid-gut; m.t. Malpighian tubule; o oesophagus; s.g. salivary gland.

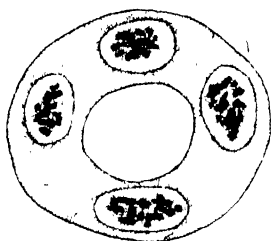
salivary gland is more elongated, its extremity reaching to two-thirds of the body-length. In the larvae of the later stages the hind-gut becomes smaller. In the fifth larval stage, the hind-gut is narrow and is divided into three parts, that is, the upper narrow part, the middle enlarged part, and the lower narrow part (Text-fig. 6, C). The walls of the mid-gut are lined with a thick epithelium. The cells composing the epithelium closely resemble each other, appearing in a large polygonal form. Each epithelial cell observed in the larva of the last stage contains two nuclei, measuring $24\text{--}36\mu$ in diameter (Text-fig. 7). The cells composing the wall of the salivary gland (Text-fig. 8) are also large, bearing nuclei measuring $15\text{--}24\mu$ in diameter. The wall of the Malpighian tubules (Text-fig. 9) consists of a single layer of epithelial cells, bearing large nuclei of $30\text{--}45\mu$ diameter.



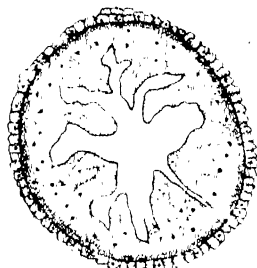
Text-fig. 7. Epithelial cells of mid-gut of the fifth stage larva. $\times 120$.



Text-fig. 9. Cross section of Malpighian tubule of the fifth stage larva. $\times 270$.



Text-fig. 8. Cross section of salivary gland of the fifth stage larva. $\times 400$.

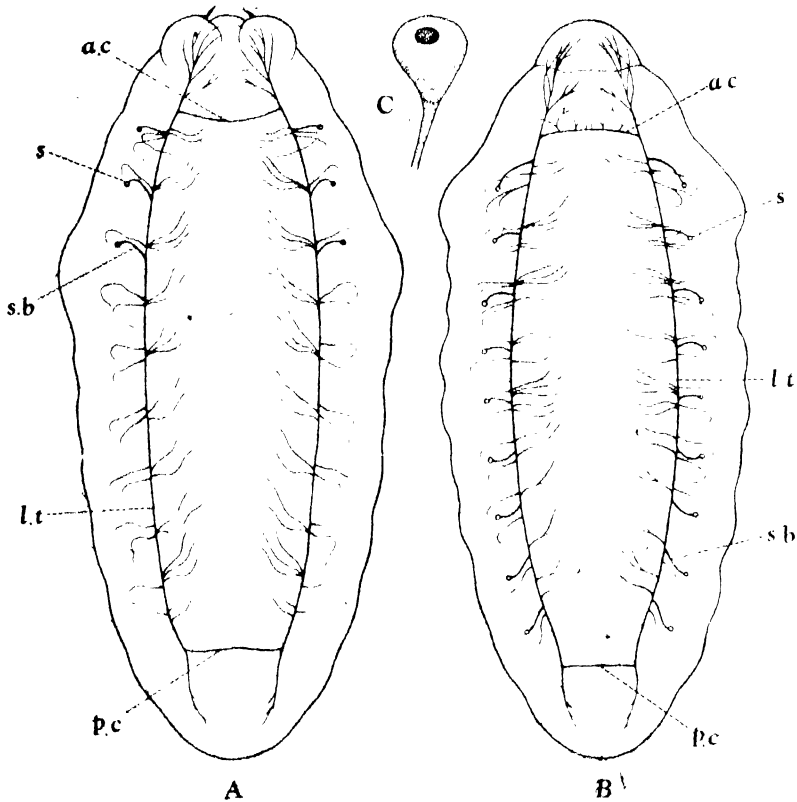


Text-fig. 10. Cross section of hind-gut of the fifth stage larva. $\times 500$.

7. Tracheal System

In the larvae of the first and the second stage, there is no indication of the tracheal system. But in the same of the later stages the tracheal system is gradually developed and becomes functional. The tracheal system of the larva of the fourth stage (Text-fig. 11, A) consists chiefly of two thread-like main lateral trunks passing along each side of the body, and uniting together anteriorly by the anterior commissure and posteriorly by the posterior commissure. Dorsal and ventral branches are given off from the main trunk in each segment of from the first to the tenth. At the anterior end of the lateral trunk several branches arise and extend towards the head. There are three pairs of open spiracles located on each segment from the second to the fourth. The tracheal system of the larva of the fifth stage (Text-fig. 11, B) is nearly similar to that of

the preceding stage, but the spiracles number nine pairs in all, opening on each segment from the second to the tenth. The spiracle leads into the spiracular trachea which connects with the main lateral trunk. The extreme end of the spiracular trachea is swollen representing a club-form structure (Text-fig. 11; C). The swollen part measures 0.02 mm. in diameter and in its center there opens a minute spiracle of 6μ diameter.



Text-fig. 11. Tracheal system of the larva. A. Fourth stage larva. $\times 50$. B. Fifth stage larva. $\times 50$. C. Spiracle of the fifth stage larva. a.c. anterior commissure; l.t. lateral trunk; p.c. posterior commissure; s. spiracle; s.b. spiracular branch.

The spiracle is always kept open and there is no apparatus to close it. In the primary larvae of the entomophagus Cynipidae hitherto known there is no indication of the tracheal system.

Many types of the primary larvae have been known among parasitic Hymenoptera. Of these, several types have been described where the

last segments are furnished with some appendages. These include for example, such forms as *Limnerium* (Ichneumonidae), *Aphidius* (Chalcidae), *Teleas* (Scelionidae) and *Eucoila* (Cynipidae). The function of the caudal appendage has been supposed by different writers to be either locomotory or respiratory. In the larva of the early stages of the entomophagous Hymenoptera whose tracheal system is apneustic, the respiration must be cutaneous, and the caudal appendage may assist the function of absorbing oxygen by increasing the body-surface. The three pairs of thoracic appendages which are characteristic to the eucoiliform larva may also assist the respiration as in the case of the caudal appendage above mentioned.

HAVILAND, in his study of *Charips*, suggests that a kind of rectal respiration might exist in the case of the larvae of the early stages. The larva of the first stage of *Charips* is heavily armoured with dark segmented plates of chitin and these hard chitinous plates must prevent the cutaneous respiration of the larva. But the hind-gut of this species encloses a large lumen lined with a layer of modified hypoderm cells, and the anus is enlarged and spined, showing a spiracle-like structure. HAVILAND states that this peculiar structure of the anus and of the hind-gut might be correlated with the respiration of the larva of this stage. It is also known that a peculiar modification of the hind-gut occurs in the case of the larvae of certain Braconids, such as *Apanteles* and *Microgaster*, that is, the body terminates in a hollow bladder or vesicle lined with hypertrophied cells. GARTENBY (1919) suggests that this structure is morphologically the hind-gut, and that it has become everted to execute the function of respiration. THORPE (1932) reports that in *Apanteles* and *Microgaster* this kind of vesicle is of great importance in executing respiration, but he also concludes by experiments that the vesicle, even when it is fully developed, it is not able to respond for more than about one-third of the total respiration. HAVILAND supposes that with regard to the respiratory function the hind-gut of *Charips* is intermediate between the highly specialized structure of the Microgastrinae and the unmodified proctodaeum of most of the Hymenopterous larvae.

In the first larval stage of *Kleidotoma japonica*, the hind-gut has a large lumen enclosed by modified hypoderm cells and the anus is of the enlarged type as in the case of *Charips*. In the second larval stage the hind-gut maintains the same globular shape seen in the first stage, although the anus is a little depressed and the spines surrounding it have disappeared. In the later stages of the larva the lumen of the hind-gut gradually diminishes in size, and is smallest of all in the full-grown larva. In both the

first and second stage of the larvae there is no indication of the tracheal system, but in the later stages it is gradually developed. In the fourth larval stage there exist three pairs of spiracles and the tracheal system becomes functional, and in the fifth stage there appear nine pairs of spiracles. The development of the tracheal system is somewhat correlated with the degeneration of the lumen of the hind-gut. The larvae of the first and second stage are floating in the body-cavity of the host-insect-larva and consequently the oxygen must be absorbed from the surrounding tissues of the same. The writer is also inclined to have the opinion that the hind-gut takes part in performing the rectal respiration. Thus oxygen may be absorbed through the hind-gut as well as from the body-surface. But the larvae of the later stages which have nearly completely devoured the pupal body of the host-insect, can take oxygen from the surrounding air by means of their spiracles. As the tracheal system becomes functional, the hind-gut may lose its function concerned with the gas-exchange and therefore it gradually degenerates.

8. Moultings and Hibernation

It is very difficult to observe directly the moultings of the larva during its development, for it grows inside the body-cavity of the host-insect and it also proves impossible to keep it alive for observation after removal from the host-insect. Therefore the determination of the number of moultings was based on the comparison of many individuals of different stages of growth, and on observations of larva which was just in the act of moulting. According to the present writer observations the larva of *Kleidotoma japonica* passes through five stages and moults four times before pupation. He was able to find many living larvae and some pupae contained within the larva and the puparia of the host-insect, at the end of October at the Tubame hot spring. At that time much snow had fallen, and the habitat of the insects seemed to be set entirely under winter conditions. From this fact it is suggested that most of *Kleidotoma japonica* probably pass the winter in the larval stage and some in the pupal stage.

VI. THE PUPA

(Pl. XXIII, Figs. 31, 32)

The pupa of *Kleidotoma japonica* (Pl. XXIII, Fig. 31) does not differ from the usual hymenopterous type, and will be described here briefly.

The female pupa measures about 2.1 mm. in length and the male about 2.0 mm. The head is nearly similar in shape to that of the adult. The antenna of the female pupa extends from the head along each side of the body, its extreme end reaching the tip of the wing, whereas the antenna of the male pupa is much longer than that of the female, its extreme end reaching the body end. The compound eye bears numerous minute depressions on its surface. The labrum is nearly a triangle in shape and its tip is protruded. The mandible is large and simple, bearing no dents. The maxillary palpi and the labial palpi are much larger than those of the adult, but their joints are scarcely recognizable (Pl. XXIII, Fig. 32).

VII. COMPARISON OF THE LARVAL CHARACTERS OF *KLEIDOTOMA JAPONICA* WITH THOSE OF OTHER ENTOMOPHAGOUS CYNIPIDAE

The biology of the entomophagous Cynipidae including the Eucoilinae, the Figitinae, and the Charipinae, has been less studied than that of most other groups of the parasitic Hymenoptera. As in the cases of some other parasitic Hymenoptera, hypermetamorphosis has been known to occur in members of the entomophagous Cynipidae, whose life-cycle has been hitherto observed. As regards the early stages of the entomophagous Cynipidae, our knowledge is limited to the following few species. KEILIN and PLUVINEL (1913) reported the life-history of *Eucoila keilini*, a parasite on *Pogomyia*, and described the larva of the first stage. This kind of larva has been termed the Eucoiliform type by later authors. HAVILAND (1921) reported the biology of the Charipinae, including *Bothryoxysta curvata*, *Charips victrix* and *Alloxysta erythrothorax*, and described their early larval forms. These flies were obtained as hyperparasites from various Aphidiidae which were primary parasites upon aphids. JAMES (1928) reported the investigations made into the life-history of four Cynipid flies, e. i. *Cothonaspis rapae* (Eucoilinae), *Kleidotoma marshalli*, *Kleidotoma* sp. (Eucoilinae), and *Figites anthomyiarum* (Figitinae), and described their larvae of the early stage. *Cothonaspis rapae* is a common parasite of the cabbage-root maggot and the other three species are reared from carrion-feeding dipterous larvae.

The primary larva of *Kleidotoma japonica* is of a typical eucoiliform bearing three pairs of thoracic appendages and a long caudal appendage. *Cothonaspis rapae* and *Kleidotoma marshalli* which belong to Eucoilinae are also known to bear the typical thoracic and caudal appendages. The

presence of the peculiar thoracic appendages and of the long caudal appendage may be taken as the distinguishing feature of the Eucoilinae. The larva of the first stage of *Charips* (Charipinae) has a long caudal appendage, but there is no indication of thoracic appendages. Furthermore, it is heavily armoured with dark segmented plates of chitin, which are not seen in other species. The larva of the first stage of *Figites anthomyiarum* resembles the typical eucoiliform type, but its thoracic appendages are very small and vestigial.

Kleidotoma japonica has a spined enlarged anus. This type of anus was first described by HAVILAND in the larva of the first stage of *Charips* and later was found by JAMES in the eucoiliform larvae of *Cothonaspis rapae* and *Kleidotoma marshalli*. KEILIN and PLUVINEL (1913), in the study of *Eucoila keilini*, did not describe the anus, having apparently omitted to examine it. The anus of *Figites anthomyiarum* is not of the enlarged type but it appears as a narrow slit-like opening. This form of anus has a somewhat similar resemblance to that possessed by the larva of the second stage of *Kleidotoma japonica*.

No mandibles were described in the case of the larva of the first stage of *Eucoila keilini* observed by KEILIN and PLUVINEL, neither in the cases of *Cothonaspis rapae*, *Kleidotoma marshalli* and *Figites anthomyiarum* investigated by JAMES. But the present writer was able to recognize clearly a pair of mandibles in the larva of the first stage of *Kleidotoma japonica*, and a beak-like sclerite situated in the middle of the mouth. HAVILAND describes a pair of slender simple mandibles in the larvae of the first and the second stage of *Charips*.

As described in the previous section, the larva of the second stage of *Kleidotoma japonica* has no any indications of thoracic appendages and the caudal appendage is very small and vestigial.

JAMES describes the larva of the second stage of each fly *Figites anthomyiarum*, *Kleidotoma marshalli* and *Kleidotoma sp.* In the case of *Figites anthomyiarum* the larva of the second stage possesses a large cephalic segment followed by eleven very clearly defined segments, to the last of which is attached a long caudal appendage. The first remarkable feature of this stage of larva is that, when it is compared with the larva of the first stage, it seems to have undergone a reduction in the number of body-segments. The second feature is that each of the first ten body-segments possesses a pair of small processes. The mouth is similar to that of the primary larva in having no mandibles. Anteriorly to the mouth the larva is armoured with a pair of long papillae, and on

the ventral surface of the head there is a conspicuous sensory organ consisting of a chitinous projection surmounted with a transparent tip. The last body-segment bears ventrally a stout caudal appendage taking a position making almost a right angle with the long axis of the body. An apneustic tracheal system is developed internally.

Many suggestions are made by JAMES with regard to the features of the early stage larvae of entomophagous Cynipidae. He notices that the larvae of the early stage bear a resemblance to certain developmental phases which, in other insects, are passed through in the egg stage. BERLESE (1913) says that there are three distinct phases in the embryology of insects, basing his argument chiefly upon the condition of the segmentation and the development of the appendages. These three are called respectively the protopod, polypod and oligopod stage. JAMES states that the larvae of the first stage of *Eucoila*, *Cothonaspis* and *Kleidotoma* correspond to the protopod embryonic stage, and hatching from the egg in these forms might occur in the middle of the same embryonic stage. As to *Figites anthomyiarum*, he places it as hatching later in embryonic development than any other first stage larva of the Cynipidae, because it is more definitely segmented. The larva of the first stage of *Charips* differs from all other larvae of the first stage of the Cynipidae in being devoid of thoracic appendages, though very reduced ones of the same appear in the second stage. JAMES considers that the larva of the second stage of *Charips* closely approximates to a protopod embryonic stage, the primary larva representing something ontogenically prior to or at the beginning of the protopod stage. He supports his opinion by the fact that the larvae of the second stage of *Figites* and *Kleidotoma*, being derived from the eucoiliform larva, represent the polypod embryonic stage. The larva of the second stage of *Figites anthomyiarum* and *Kleidotoma marshalli* illustrated by JAMES, exactly resemble the polypod embryonic stage in possessing a pair of short appendages on each body-segment. Accordingly JAMES terms this type of larva the polypodeiform. He is also of the opinion that a great number of the parasitic Cynipidae which possess the definite protopod stage of primary larvae, will be found to have polypod stages in addition.

The larva of the second stage of *Kleidotoma japonica* is different from those of *Figites anthomyiarum* and *Kleidotoma marshalli* which are described by JAMES on account of the absence of the segmental appendages and of the long caudal appendage, though its larva of the first stage may be of the typical eucoiliform. The short processes which mainly characterize the larva of the polypodeiform type, could not be observed

in any body-segment of *Kleidotoma japonica*. This fact does not agree with the conclusion come to by JAMES.

In comparing the larvae of the last stage of the various insects above observed, we find that there exist certain structural differences among them. The full-grown larvae of *Kleidotoma japonica*, *Figites anthomyiarum* (BOUCHÉ, 1834; JAMES, 1928), and *Eucoila keilini* (KEILIN and PLUVINEL, 1913) possess twelve segments, whereas those of *Charips* (HAVILAND, 1923) and of *Anacharis typica* (HANDLIRSCH, 1886) have thirteen. As to the number of spiracles, the full-grown larvae of *Kleidotoma japonica*, *Kleidotoma keilini* and *Figites anthomyiarum* are furnished with nine pairs, while *Cothonaspis rapae* has eight pairs and *Charips* possesses only six pairs.

VIII. SUMMARY

1) The cynipid fly which is described in this paper is new to science and is named *Kleidotoma japonica*.

2) *Kleidotoma japonica* is an entomophagous parasite to be found inside the larva and the pupa of the Hot Spring Fly, *Scatella calida* MATSUMURA.

3) The larvae of the Hot Spring Fly live in the waters of hot springs which range from 30° to 40°C in temperature and from 4.0 to 6.5 in pH.

4) When ready to emerge, the adult of *Kleidotoma japonica* pierces a hole in the dorsal side of the puparium of the host-insect, and then creeps out from it. The hole thus made is nearly round in shape, with a diameter of about 0.7 mm.

5) The egg of *K. japonica* is deposited inside the body-cavity of the larva of the first or of the second stage of the host. The egg laid is an elongated oval in shape, measuring 0.42 mm. in length and 0.12 mm. in breadth. It bears at its anterior end a narrow peduncle 0.25 mm. long.

6) Hypermetamorphosis occurs in the larval history of *K. japonica* as in other cases of entomophagous Cynipidae hitherto known.

a) The larva of the first stage is of the typical eucoiliform provided with three pairs of thoracic appendages together with a long caudal appendage. The mouth is of suctorial type, projecting on the ventral surface of the head, and is furnished with a pair of mandibles and with a beak-like sclerite. The anus is large and is of spined type, opening near the posterior end of the body, and has a diameter of 0.038-0.045 mm.

b) The larva of the second stage is devoid of the thoracic appendages and of the long caudal appendage. The body is cylindrical in form, the

anterior end being more or less truncated and the posterior end pointed. The mandibles are large and thick, and the maxillae are fused together with the labium. The anus is transversely oval in shape and bears no spines.

c) The larva of the third stage is an elongated oval in body-shape, and with the posterior end less pointed than in the case of the preceding stage. The head has a pair of slight protuberances of area antennalis.

d) The larva of the fourth stage is grub-like in form and is furnished with three pairs of spiracles. The mandibles are strongly chitinized and are sharply pointed at the apex.

e) The larva of the fifth stage is in the form of a legless grub of the usual hymenopterous type, provided with nine pairs of spiracles. The mandible is strongly chitinized and is dark brown in colour, bearing about ten dents.

7) In the larvae both of the first and the second stages there are no indications of a tracheal system. The larvae of both the fourth and fifth stages have a well-developed tracheal system. It consists chiefly of two main trunks, which pass along each side of the body, and are connected with each other by the anterior and posterior commissures. The larva of the fourth stage has three pairs of spiracles, but that of the fifth stage has nine pairs.

8) The digestive canal is divided into three portions, — the fore-gut, the mid-gut and the hind-gut. The mid-gut forms a voluminous sack occupying the greater part of the body-cavity. The hind-gut of the larva of the first and the second stage has a large globular lumen inside which opens into the enlarged anus. It is very likely that the hind-gut of these stages assists the act of absorption of oxygen from the host's tissues and thus there may exist a kind of rectal respiration in the larvae of early stages. The hind-gut seen in the later stages proportionally diminishes in dimensions and does not appear globular in form. There are a pair of salivary glands and a pair of Malpighian tubules on the ventral side of the guts.

9) The full-grown larva devours the pupal-body of the host completely, and then pupates within the puparium of the same.

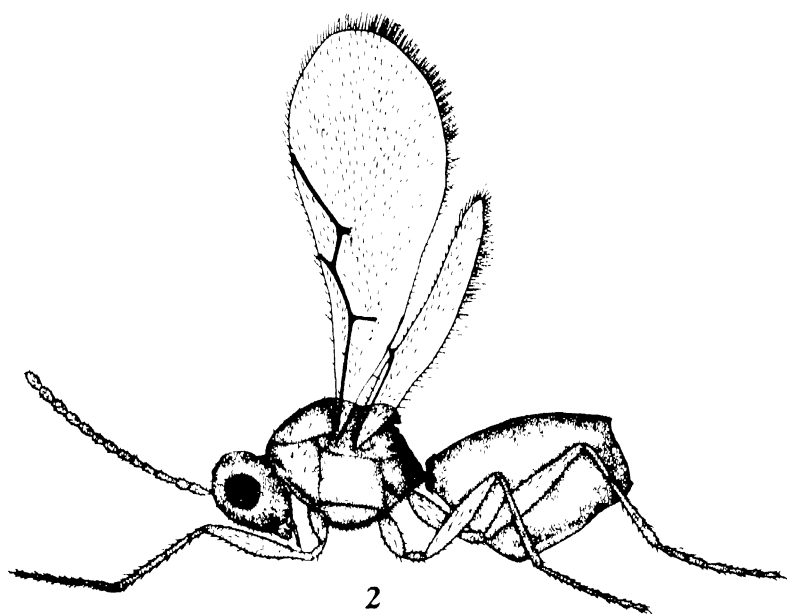
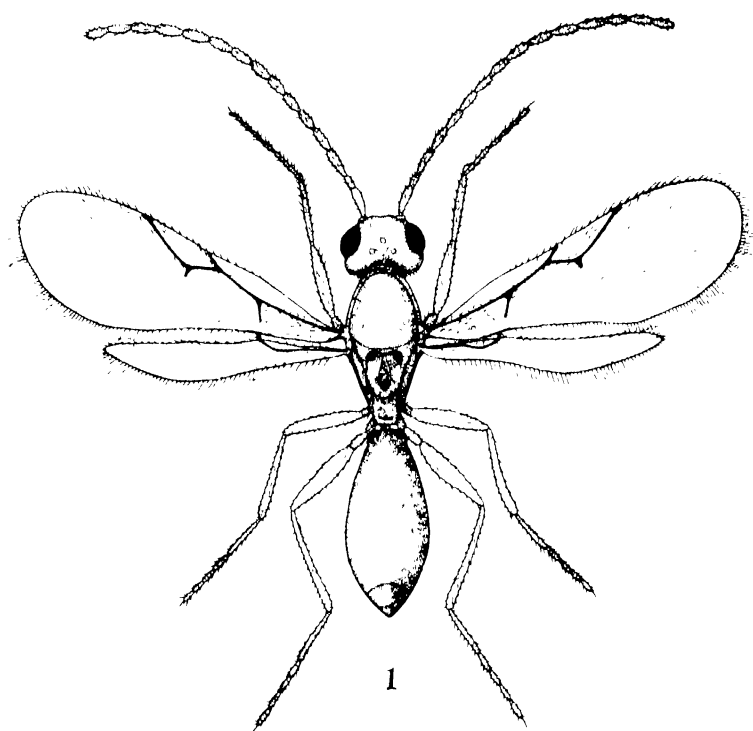
10) The larva of the early stage of *Kleidotoma japonica*, n. sp. is compared with those of the other entomophagous Cynipidae hitherto known and is discussed as regards its structure.

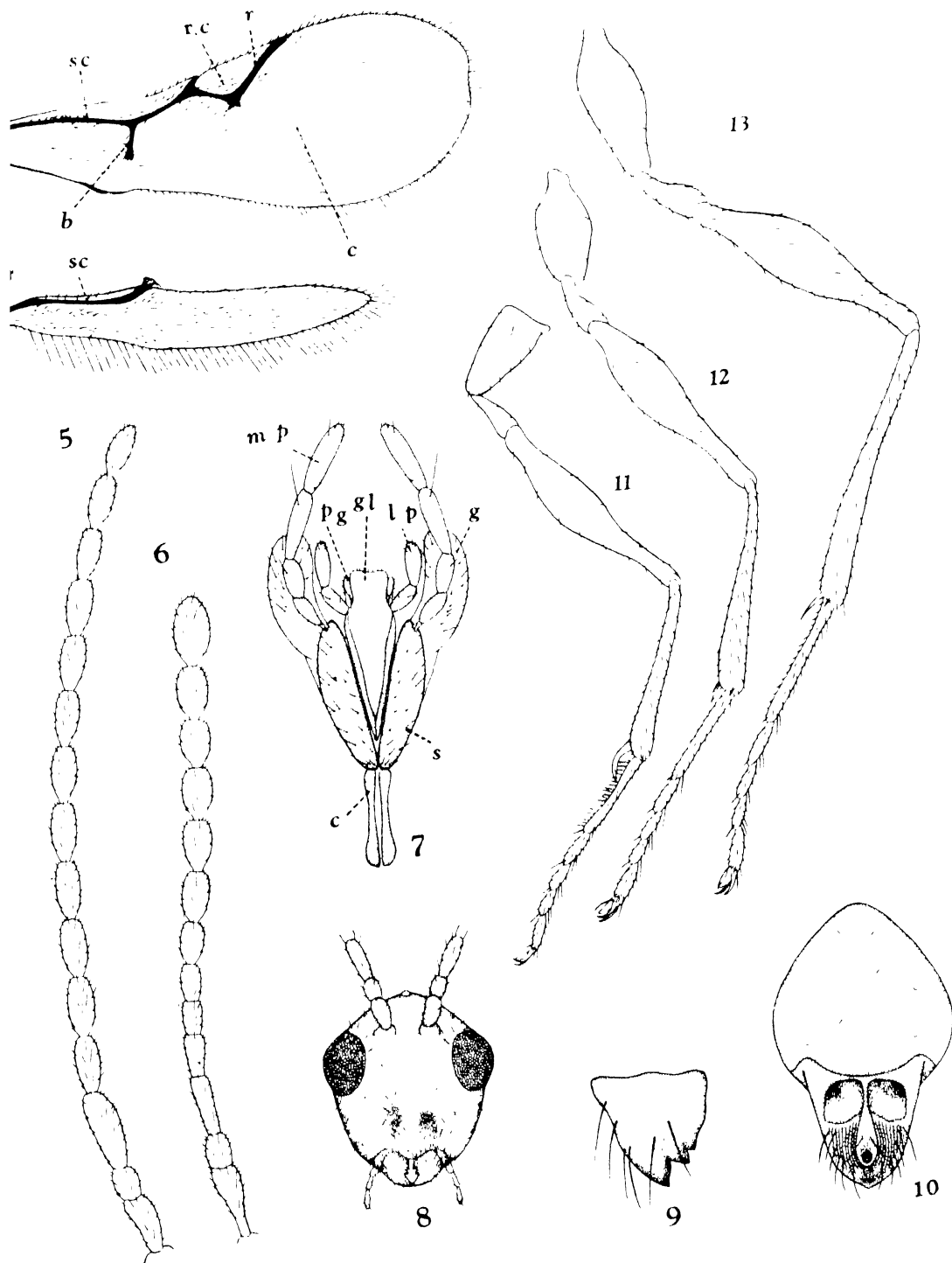
IX. LITERATURE CITED

- ADLER, H. (1881). Über den Generationswechsel der Eichen-Gallwespen. Zeit. Wiss. Zool. Bd. 35.
- CAMLON, P. (1890). A Monograph of the British Phytophagous Hymenoptera. Ray Society's Publication, Vol. 3.
- HAVILAND, M. D. (1921). Preliminary Note on a Cynipid Hyperparasite of Aphides. Proc. Phil. Soc. Camb., Vol. 20, No. 2.
- (1921). On the Bionomics and Post-Embryonic Development of Certain Cynipid Hyperparasites of Aphids. Quart. Journ. Mic. Sci. Vol. 65.
- JAMES, H. C. (1928). On the Life Histories and Economic States of Certain Cynipid Parasites of Dipterous Larvae, with Descriptions of Some New Larval Forms. Ann. Appl. Biol., Vol. 15, No. 2.
- KIEFFER, J. J. and DAHL TORRE, K. W. (1910). "Cynipidae" in Das Tierreich. Berlin.
- KILIN, D. et BAUMI PRUVINEL, C. (1913). Formes Larvaires et Biologie d'un Cynipide Entomophage. Bull. Sci. France, Septieme série Tome 17.
- KIRMAYER, R. (1909). Bau und Entwicklung der Mundteil bei *Vespa vulgaris*. Morph. Jahrb., Bd. 39.
- NELSON, J. A. (1920). Morphology of the Honey Bee Larva. Journ. Agr. Res., Vol. 28, No. 12.
- MARSHALL, P. (1906). Les Platygasters. Arch. Zool. Expér., Quatorzieme Série Tome 4.
- MATSUMURA, S. (1915). On a Fly living in Hot Spring (Japanese). Insect World, Vol. 19, No. 6.
- OKADA, Y. (1936). On the Biological Studies of Thermal Waters in Japan (I). Rep. Jap. Association for Adv. Sci., Vol. 11, No. 3.
- OKADA, Y., UYEMURA, M. and ITO, Y. (1937). On the Biological Studies of Thermal Waters in Japan (IX). The Fauna of Tubame and Seki Hot Springs at the Foot of Mt. Myoko, Nügata-ken. (Japanese). Zool. Mag., Vol. 49, No. 12.
- PAKER, H. L. (1924). Recherches sur les Formes Post-embryonnaires des Chalcidiens. Ann. Soc. Ent. Fr., Tome 93.
- RICHARDSON, C. H. (1913). Studies on the Habits and Development of a Hymenopterous Parasite, *Spalangia muscidarum* RICHARDSON. Journ. Morph., Vol. 24.
- THORPE, W. H. (1932). Experiments upon Respiration in the Larvae of Certain Parasitic Hymenoptera. Proc. Royal Soc. London, Ser. B, Vol. 109.

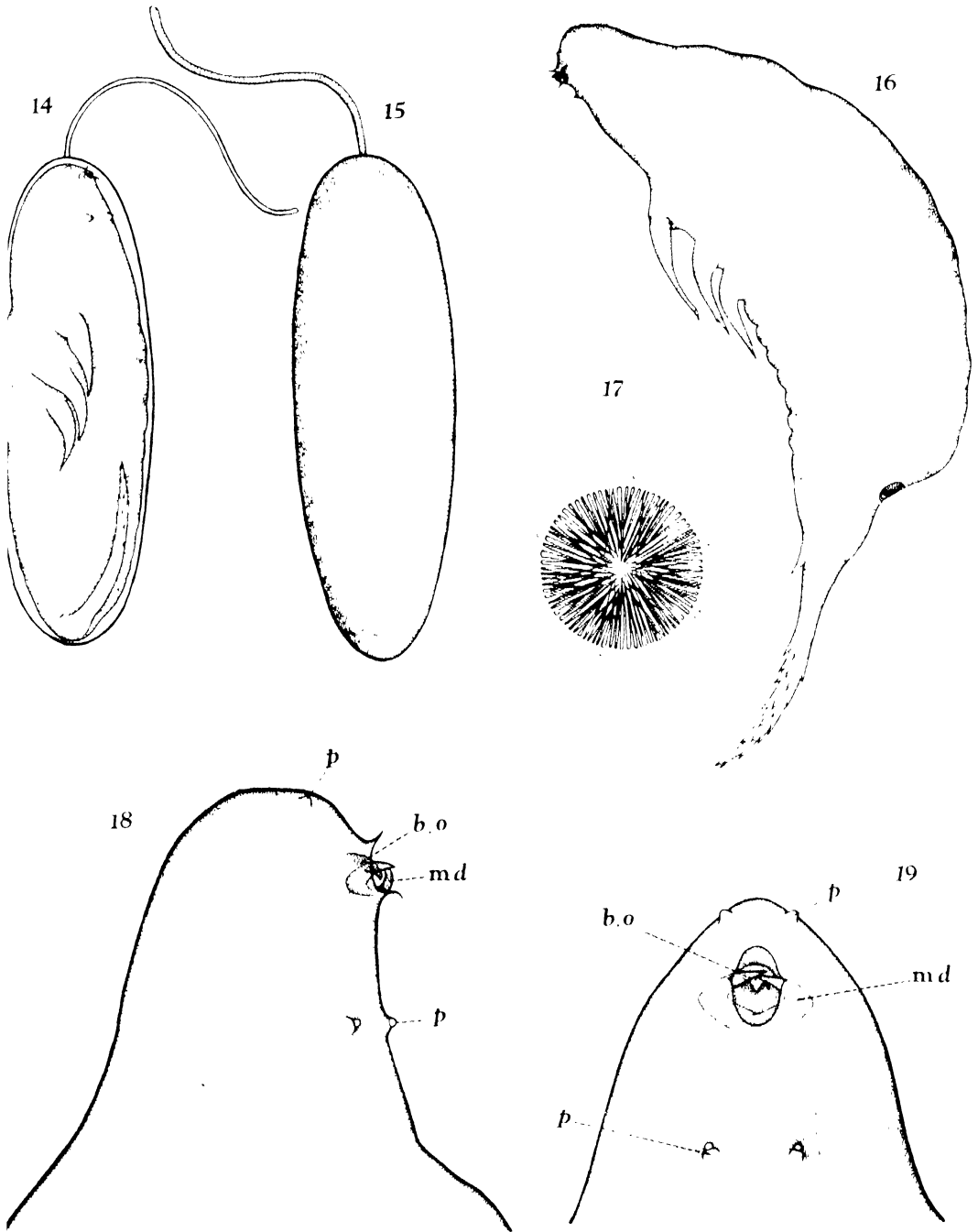
EXPLANATIONS OF PLATES

- Plate XIX. Fig. 1. *Kleidotoma japonica*, n. sp. ♂ ca. $\times 30$.
 Fig. 2. *Kleidotoma japonica*, n. sp. ♀ ca. $\times 30$.
- Plate XX. Fig. 3. Fore wing of female. $\times 50$.
 Fig. 4. Hind wing of the same. $\times 50$.
 Sc subcosta; b basalis; r radius; c cubitus; r.c radial cellule
 Fig. 5. Antenna of male. $\times 65$.
 Fig. 6. Antenna of female. $\times 65$.
 Fig. 7. Maxillae and labium of female. $\times 200$.
 c cardo; g galea; gl glossa; lp labial palpus; m.p. maxillary palpus; p paraglossa; s stipes
 Fig. 8. Head of female, frontal view. $\times 66$.
 Fig. 9. Mandible of the same. $\times 160$.
 Fig. 10. Thorax of female, dorsal view. $\times 66$.
 Fig. 11. Fore leg of female. $\times 66$.
 Fig. 12. Mid leg of the same. $\times 66$.
 Fig. 13. Hind leg of the same. $\times 66$.
- Plate XXI. Fig. 14. Egg, just before hatching out. $\times 200$.
 Fig. 15. Egg.
 Fig. 16. First stage larva. $\times 200$.
 Fig. 17. Anus of the same. $\times 660$.
 Fig. 18. Head of the same, lateral view. $\times 660$.
 Fig. 19. The same, frontal view. $\times 660$.
 b.s beak-like sclerite; md mandible; p papilla
- Plate XXII. Fig. 20. Second stage larva, ventral view. $\times 130$.
 Fig. 21. Head of the same, frontal view. $\times 270$.
 Fig. 22. Posterior part of the same, dorsal view. $\times 200$.
 a anus
 Fig. 23. Third stage larva, dorsal view. $\times 40$.
 Fig. 24. Head of the same, dorsal view. $\times 170$.
 a.a area antennalis; l labium; la labrum; md mandible; mx maxilla
- Plate XXIII. Fig. 25. Fourth stage larva, dorsal view. $\times 27$.
 Fig. 26. Head of the same, frontal view. $\times 170$.
 a.a area antennalis; l labium; la labrum; md mandible; mx maxilla
 Fig. 27. Fifth stage larva, lateral view. $\times 27$.
 Fig. 28. Mandible of the same. $\times 660$.
 Fig. 29. Mouth-parts of the same.
 l labium; la labrum; md mandible; mx maxilla
 Fig. 30. Fifth stage larva, lateral view. $\times 27$.
 Fig. 31. Pupa, lateral view. $\times 50$.
 Fig. 32. Head of the same. $\times 66$.
 cl clypeus; la labium; md mandible; mx maxilla; m.p maxillary palpus; lp labial palpus; g glossa

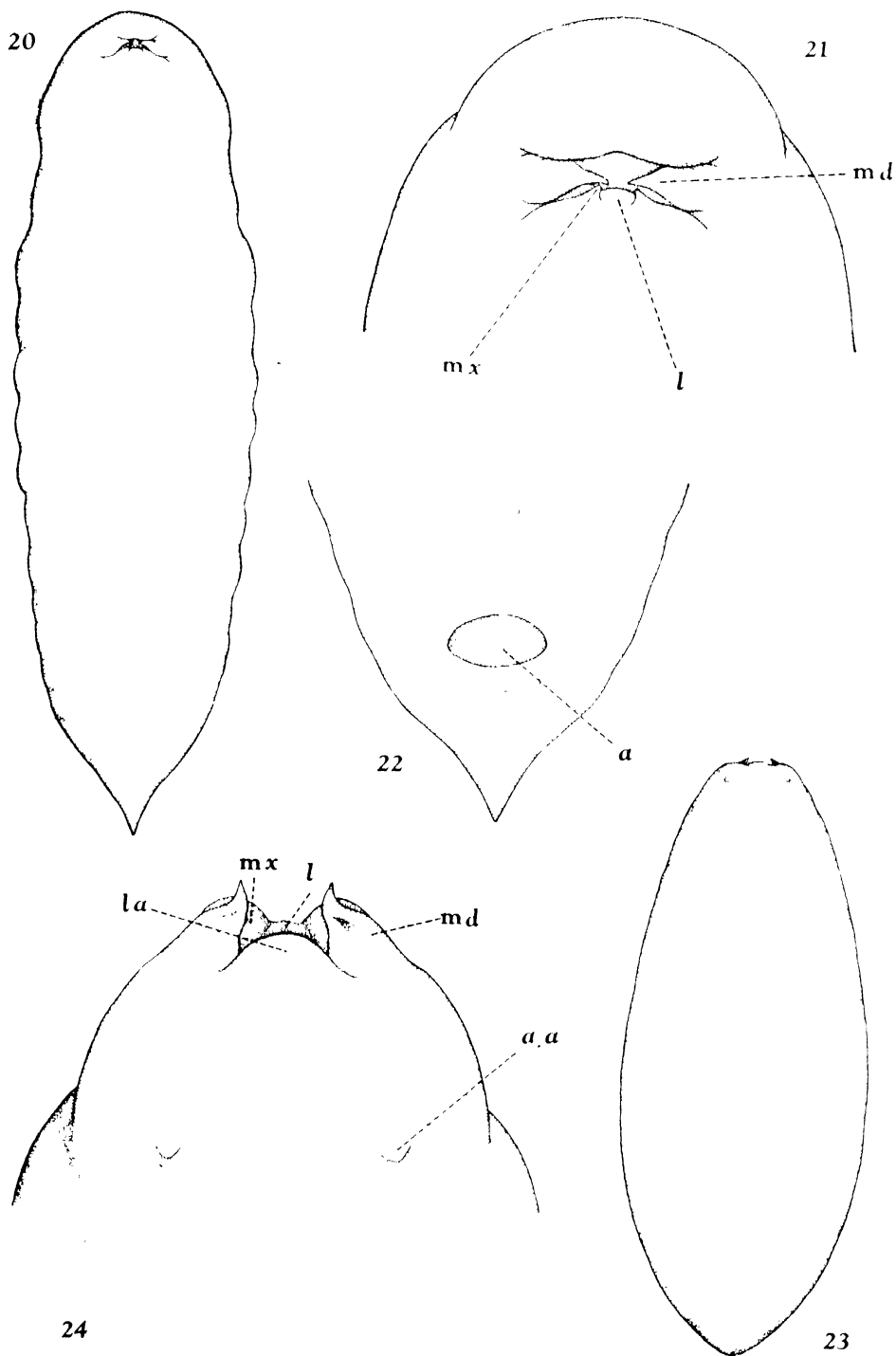


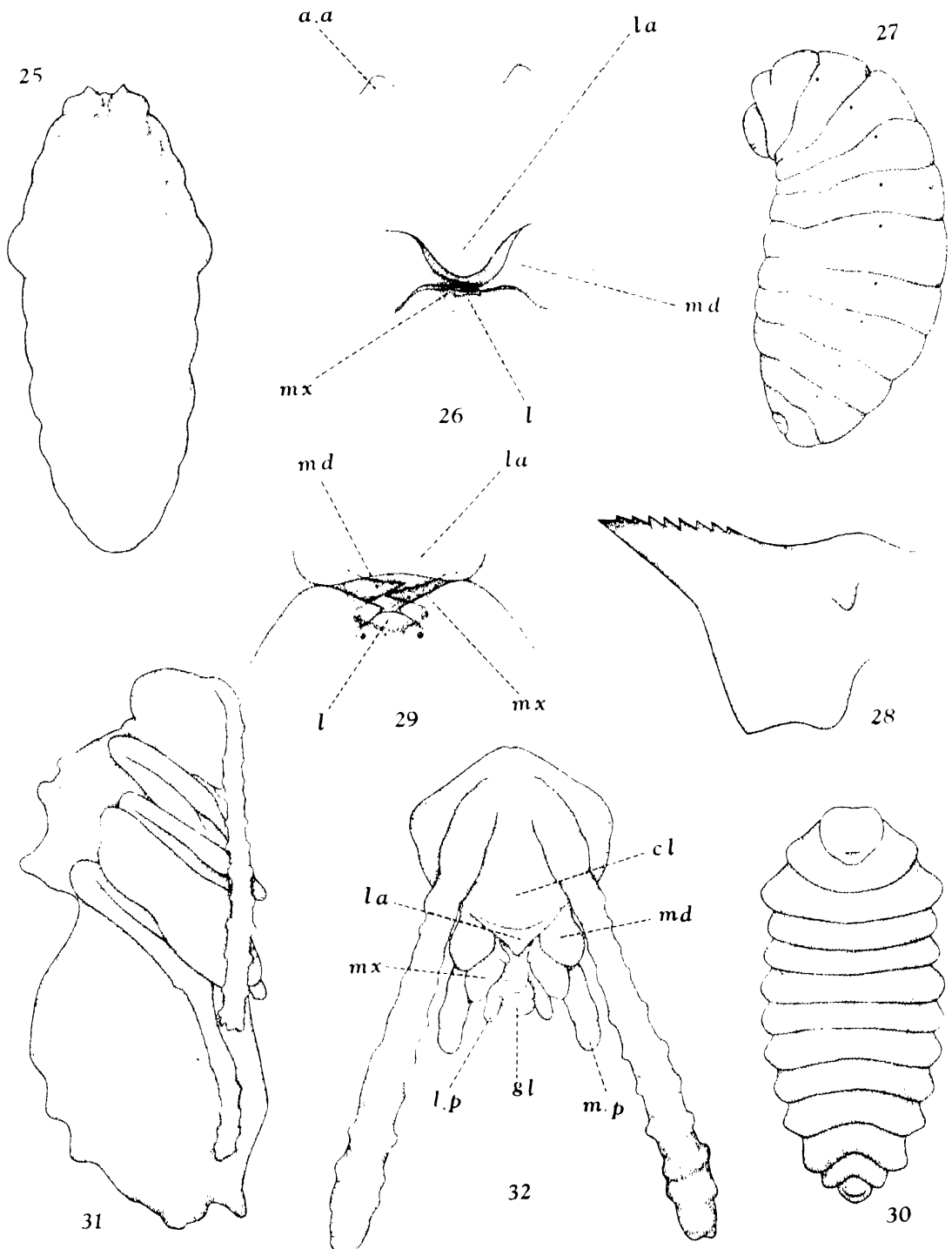


K. HUZIMATU: The life history of a new Cynipid fly.



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I. A. R. I. 75.

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